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#### (57) Abstract

The present invention relates generally to isolated genes which encode polypeptides involved in cellulose biosynthesis in plants and transgenic plants expressing same in sense or antisense orientation, or as ribozymes, co-suppression or gene-targeting molecules. More particularly, the present invention is directed to a nucleic acid molecule isolated from Arabidopsis thaliana, Oryza sativa, wheat, barley, maize, Brassica ssp., Gossypium hirsutum and Eucahyptus ssp. which encode an enzyme which is important in cellulose biosynthesis, in particular the cellulose synthase enzyme and homologues, analogues and derivatives thereof and uses of same in the production of transgenic plants expressing altered cellulose biosynthetic properties.

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### "MANIPULATION OF CELLULOSE AND/OR β-1,4-GLUCAN"

The present invention relates generally to isolated genes which encode polypeptides involved in cellulose biosynthesis and transgenic organisms expressing same in sense or antisense or orientation, or as ribozymes, co-suppression or gene-targeting molecules. More particularly, the present invention is directed to a nucleic acid molecule isolated from Arabidopsis thaliana, Oryza sativa, wheat, barley, maize, Brassica ssp., Gossypium hirsutum and Eucalyptus ssp. which encode an enzyme which is important in cellulose biosynthesis, in particular the cellulose synthase enzyme and homologues, analogues and derivatives thereof and uses of same in the production of transgenic plants expressing altered cellulose biosynthetic properties.

Bibliographic details of the publications referred to by author in this specification are collected at the end of the description. Sequence identity numbers (SEQ ID Nos.) for the nucleotide and amino acid sequences referred to in the specification are defined after the bibliography.

Throughout the specification, unless the context requires otherwise, the word "comprise", or variations such as "comprises" or "comprising" will be understood to imply the inclusion of 20 a stated element or integer or group of elements or integers but not the exclusion of any other element or integer or group of elements or integers.

Cellulose, the world's most abundant biopolymer, is the most characteristic component of plant cell walls in so far as it forms much of the structural framework of the cell wall.

25 Cellulose is comprised of crystalline β-1,4-glucan microfibrils. The crystalline microfibrils are extremely strong and resist enzymic and mechanical degradation, an important factor in determining the nutritional quantity, digestibility and palatability of animal and human foodstuffs. As cellulose is also the dominant structural component of industrially-important plant fibres, such as cotton, flax, hemp, jute and the timber crops such as Eucalyptus ssp. and

30 Pinus ssp., amongst others, there is considerable economic benefit to be derived from the

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manipulation of cellulose content and/or quantity in plants. In particular, the production of food and fibre crops with altered cellulose content are highly desirable objectives.

The synthesis of cellulose involves the  $\beta$ -1,4-linkage of glucose monomers, in the form of a nucleoside diphospoglucose such as UDP-glucose, to a pre-existing cellulose chain, catalysed by the enzyme cellulose synthase.

Several attempts to identify the components of the functional cellulose synthase in plants have failed, because levels of β-1,4-glucan or crystalline cellulose produced in such assays have 10 hitherto been too low to permit enzyme purification for protein sequence determination. Insufficient homology between bacterial β-1,4-glucan synthase genes and plant cellulose synthase genes has also prevented the use of hybridisation as an approach to isolating the plant homologues of bacterial β-1,4-glucan (cellulose) synthases.

15 Furthermore, it has not been possible to demonstrate that the cellulose synthase enzyme from plants is the same as, or functionally related to, other purified and characterised enzymes involved in polysaccharide biosynthesis. As a consequence, the cellulose synthase enzyme has not been isolated from plants and, until the present invention, no nucleic acid molecule has been characterised which functionally-encodes a plant cellulose synthase enzyme.

20

In work leading up to the present invention, the inventors have generated several novel mutant Arabidopsis thaliana plants which are defective in cellulose biosynthesis. The inventors have further isolated a cellulose synthase gene designated RSW1, which is involved in cellulose biosynthesis in Arabidopsis thaliana, and homologous sequences in Oryza sativa, wheat, barley, maize, Brassica ssp., Gossypium hirsutum and Eucalyptus ssp. The isolated mucleic acid molecules of the present invention provide the means by which cellulose content and structure may be modified in plants to produce a range of useful fibres suitable for specific industrial purposes, for example increased decay resistance of timber and altered digestibility of foodstuffs, amongst others.

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Accordingly, one aspect of the present invention provides an isolated nucleic acid molecule comprising a sequence of nucleotides which encodes, or is complementary to a sequence which encodes a polypeptide of the cellulose biosynthetic pathway or a functional homologue, analogue or derivative thereof.

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The nucleic acid molecule of the invention may be derived from a prokaryotic source or a eukaryotic source.

Those skilled in the art will be aware that cellulose production requires not only the presence of a catalytic subunit, but also its activation and organisation into arrays which favour the crystallization of glucan chains. This organisation is radically different between bacteria, which possess linear arrays, and higher plants, which possess hexameric clusters or "rosettes", of glucan chains. The correct organisation and activation of the bacterial enzyme may require many factors which are either not known, or alternatively, not known to be present in plant cells, for example specific membrane lipids to impart an active conformation on the enzyme complex or protein, or the bacterial c-di-GMP activation system. Accordingly, the use of a plant-derived sequence in eukaryotic cells such as plants provides significant advantages compared to the use of bacterially-derived sequences.

- 20 Accordingly, the present invention does not extend to known genes encoding the catalytic subunit of Agrobacterium tumefaciens or Acetobacter xylinum or Acetobacter pasteurianus cellulose synthase, or the use of such known bacterial genes and polypeptides to manipulate cellulose.
- 25 Preferably, the subject nucleic acid molecule is derived from an eukaryotic organism.

In a more preferred embodiment of the invention, the isolated nucleic acid molecule of the invention encodes a plant cellulose synthase or a catalytic subunit thereof, or a homologue, analogue or derivative thereof.

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More preferably, the isolated nucleic acid molecule encodes a plant cellulose synthase polypeptide which is associated with the primary cell wall of a plant cell. In an alternative preferred embodiment, the nucleic acid molecule of the invention encodes a plant cellulose synthase or catalytic subunit thereof which is normally associated with the secondary cell wall 5 of a plant cell.

In a more preferred embodiment, the nucleic acid molecule of the invention is a cDNA molecule, genomic clone, mRNA molecule or a synthetic oligonucleotide molecule.

In a particularly preferred embodiment, the present invention provides an isolated nucleic acid molecule which encodes or is complementary to a nucleic acid molecule which encodes the *Arabidopsis thaliana*, *Gossypium hirsutum* (cotton), *Oryza sativa* (rice), *Eucalyptus ssp.*, *Brassica ssp.* wheat, barley or maize cellulose synthase enzyme or a catalytic subunit thereof or a polypeptide component, homologue, analogue or derivative thereof.

15

As exemplified herein, the present inventors have identified cellulose biosynthesis genes in maize, wheat, barley, rice, cotton, *Brassica ssp.* and *Eucalyptus ssp.*, in addition to the specific *Arabidopsis thaliana RSW*1 gene sequence which has been shown to be particularly useful for altering cellulose and/or  $\beta$ -1,4-glucan and/or starch levels in cells.

20

Hereinafter the term "polypeptide of the cellulose biosynthetic pathway" or similar term shall be taken to refer to a polypeptide or a protein or a part, homologue, analogue or derivative thereof which is involved in one or more of the biosynthetic steps leading to the production of cellulose or any related β-1,4-glucan polymer in plants. In the present context, a polypeptide of the cellulose biosynthetic pathway shall also be taken to include both an active enzyme which contributes to the biosynthesis of cellulose or any related β-1,4-glucan polymer in plants and to a polypeptide component of such an enzyme. As used herein, a polypeptide of the cellulose biosynthetic pathway thus includes cellulose synthase. Those skilled in the art will be aware of other cellulose biosynthetic pathway polypeptides in plants.

The term "related β-1,4-glucan polymer" shall be taken to include any carbohydrate molecule comprised of a primary structure of β-1,4-linked glucose monomers similar to the structure of the components of the cellulose microfibril, wherein the relative arrangement or relative configuration of the glucan chains may differ from their relative configuration in microfibrils of cellulose. As used herein, a related β-1,4-glucan polymer includes those β-1,4-glucan polymers wherein individual β-1,4-glucan microfibrils are arranged in an anti-parallel or some other relative configuration not found in a cellulose molecule of plants and those non-crystalline β-1,4-glucans described as lacking the resistance to extraction and degradation that characterise cellulose microfibrils.

10

The term "cellulose synthase" shall be taken to refer to a polypeptide which is required to catalyse a  $\beta$ -1.4-glucan linkage to a cellulose microfibril.

Reference herein to "gene" is to be taken in its broadest context and includes:

- 15 (i) a classical genomic gene consisting of transcriptional and/or translational regulatory sequences and/or a coding region and/or non-translated sequences (i.e. introns, 5'- and 3'- untranslated sequences); or
  - (ii) mRNA or cDNA corresponding to the coding regions (i.e. exons) and 5'- and 3'- untranslated sequences of the gene.

20

The term "gene" is also used to describe synthetic or fusion molecules encoding all or part of a functional product.

In the present context, the term "cellulose gene" or "cellulose genetic sequence" or similar term shall be taken to refer to any gene as hereinbefore defined which encodes a polypeptide of the cellulose biosynthetic pathway and includes a cellulose synthase gene.

The term "cellulose synthase gene" shall be taken to refer to any cellulose gene which specifically encodes a polypeptide which is a component of a functional enzyme having 30 cellulose synthase activity i.e. an enzyme which catalyses a β-1,4-glucan linkage to a

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cellulose microfibril.

Preferred cellulose genes may be derived from a naturally-occurring cellulose gene by standard recombinant techniques. Generally, a cellulose gene may be subjected to 5 mutagenesis to produce single or multiple nucleotide substitutions, deletions and/or additions. Nucleotide insertional derivatives of the cellulose synthase gene of the present invention include 5' and 3' terminal fusions as well as intra-sequence insertions of single or multiple nucleotides. Insertional nucleotide sequence variants are those in which one or more nucleotides are introduced into a predetermined site in the nucleotide sequence although random insertion is also possible with suitable screening of the resulting product. Deletional variants are characterised by the removal of one or more nucleotides from the sequence. Substitutional nucleotide variants are those in which at least one nucleotide in the sequence has been removed and a different nucleotide inserted in its place. Such a substitution may be "silent" in that the substitution does not change the amino acid defined by the codon.

15 Alternatively, substituents are designed to alter one amino acid for another similar acting amino acid, or amino acid of like charge, polarity, or hydrophobicity.

As used herein, the term "derived from" shall be taken to indicate that a particular integer or group of integers has originated from the species specified, but has not necessarily been 20 obtained directly from the specified source.

For the present purpose, "homologues" of a nucleotide sequence shall be taken to refer to an isolated nucleic acid molecule which is substantially the same as the nucleic acid molecule of the present invention or its complementary nucleotide sequence, notwithstanding the occurrence within said sequence, of one or more nucleotide substitutions, insertions, deletions, or rearrangements.

"Analogues" of a nucleotide sequence set forth herein shall be taken to refer to an isolated nucleic acid molecule which is substantially the same as a nucleic acid molecule of the present invention or its complementary nucleotide sequence, notwithstanding the occurrence of any

non-nucleotide constituents not normally present in said isolated nucleic acid molecule, for example carbohydrates, radiochemicals including radionucleotides, reporter molecules such as, but not limited to DIG, alkaline phosphatase or horseradish peroxidase, amongst others.

5 "Derivatives" of a nucleotide sequence set forth herein shall be taken to refer to any isolated nucleic acid molecule which contains significant sequence similarity to said sequence or a part thereof. Generally, the nucleotide sequence of the present invention may be subjected to mutagenesis to produce single or multiple nucleotide substitutions, deletions and/or insertions. Nucleotide insertional derivatives of the nucleotide sequence of the present invention include 5° and 3° terminal fusions as well as intra-sequence insertions of single or multiple nucleotides or nucleotide analogues. Insertional nucleotide sequence variants are those in which one or more nucleotides or nucleotide analogues are introduced into a predetermined site in the nucleotide sequence of said sequence, although random insertion is also possible with suitable screening of the resulting product being performed. Deletional variants are characterised by the removal of one or more nucleotides from the nucleotide sequence. Substitutional nucleotide variants are those in which at least one nucleotide in the sequence has been removed and a different nucleotide or nucleotide analogue inserted in its place.

The present invention extends to the isolated nucleic acid molecule when integrated into the genome of a cell as an addition to the endogenous cellular complement of cellulose synthase genes. The said integrated nucleic acid molecule may, or may not, contain promoter sequences to regulate expression of the subject genetic sequence.

The isolated nucleic acid molecule of the present invention may be introduced into and 25 expressed in any cell, for example a plant cell, fungal cell, insect cell, animal cell, yeast cell or bacterial cell. Those skilled in the art will be aware of any moficiations which are required to the codon usage or promoter sequences or other regulatory sequences, in order for expression to occur in such cells.

30 Another aspect of the present invention is directed to a nucleic acid molecule which comprises

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a sequence of nucleotides corresponding or complementary to any one or more of the sequences set forth in SEQ ID Nos:1, 3, 4, 5, 7, 9, 11, or 13, or having at least about 40%, more preferably at least about 55%, still more preferably at least about 65%, yet still more preferably at least about 75-80% and even still more preferably at least about 85-95% nucleotide similarity to all, or a part thereof.

According to this aspect of the invention, said nucleic acid molecule encodes, or is complementary to a nucleotide sequence encoding, a polypeptide of the cellulose biosynthetic pathway in a plant or a homologue, analogue or derivative thereof.

10

Preferably, a nucleic acid molecule which is at least 40% related to any one or more of the sequences set forth in SEQ ID Nos:1, 3, 4, 5, 7, 9, 11, or 13 comprises a nucleotide sequence which encodes or is complementary to a sequence which encodes a plant cellulose synthase, more preferably a cellulose synthase which is associated with the primary or the 15 secondary plant cell wall of the species from which it has been derived.

Furthermore, the nucleic acid molecule according to this aspect of the invention may be derived from a monocotyledonous or dicotyledonous plant species. In a particularly preferred embodiment, the nucleic acid molecule is derived from *Arabidopsis thaliana*, *Oryza sativa*, 20 wheat, barley, maize, *Brassica ssp.*, *Gossypium hirsutum* (cotton) or *Eucalyptus ssp.*, amongst others.

For the purposes of nomenclature, the nucleotide sequence shown in SEQ ID NO:1 relates to a cellulose gene as hereinbefore defined which comprises a cDNA sequence designated T20782 and which is derived from *Arabidopsis thaliana*. The amino acid sequence set forth in SEQ ID NO:2 relates to the polypeptide encoded by T20782.

The nucleotide sequence set forth in SEQ ID NO:3 relates to the nucleotide sequence of the complete *Arabidopsis thaliana* genomic gene *RSW*1, including both intron and exon 30 sequences. The nucleotide sequence of SEQ ID NO:3 comprises exons 1-14 of the genomic

gene and includes 2295bp of 5'-untranslated sequences, of which approximately the first 1.9kb comprises RSW1 promoter sequence (there is a putative TATA box motif at positions 1843-1850 of SEQ ID NO:3). The nucleotide sequence set forth in SEQ ID NO:3 is derived from the cosmid clone 23H12. This sequence is also the genomic gene equivalent of SEQ ID No:1 and 5.

The nucleotide sequence set forth in SEQ ID NO:4 relates to the partial nucleotide sequence of a genomic gene variant of RSW1, derived from cosmid clone 12C4. The nucleotide sequence of SEQ ID NO:4 comprises exon sequence 1-11 and part of exon 12 of the genomic gene sequence and includes 862bp of 5'-untranslated sequences, of which approximately 700 nucleotides comprise RSW1 promoter sequences (there is a putative TATA box motif at positions 668-673 of SEQ ID NO:4). The genomic gene sequence set forth in SEQ ID NO:4 is the equivalent of the cDNA sequence set forth in SEQ ID NO:7 (i.e. cDNA clone Ath-A).

15 The nucleotide sequence shown in SEQ ID NO:5 relates to a cellulose gene as hereinbefore defined which comprises a cDNA equivalent of the *Arabidopsis thaliana RSW*1 gene set forth in SEQ ID NO:3. The amino acid sequence set forth in SEQ ID NO:6 relates to the polypeptide encoded by the wild-type *RSW*1 gene sequences set forth in SEQ ID Nos:3 and 5.

20

The nucleotide sequence shown in SEQ ID NO:7 relates to a cellulose gene as hereinbefore defined which comprises a cDNA equivalent of the *Arabidopsis thaliana RSW*1 gene set forth in SEQ ID NO:4. The nucleotide sequence is a variant of the nucleotide sequences set forth in SEQ ID Nos:3 and 5. The amino acid sequence set forth in SEQ ID NO:8 relates to the polypeptide encoded by the wild-type *RSW*1 gene sequences set forth in SEQ ID Nos:4 and 6.

The nucleotide sequence shown in SEQ ID NO:9 relates to a cellulose gene as hereinbefore defined which comprises a further wild-type variant of the *Arabidopsis thaliana RSW*1 gene 30 set forth in SEQ ID Nos:3 and 5. The nucleotide sequence variant is designated *Ath-B*. The

amino acid sequence set forth in SEQ ID NO:10 relates to the polypeptide encoded by the wild-type RSW1 gene sequence set forth in SEQ ID No:9.

The nucleotide sequence shown in SEQ ID NO:11 relates to a cellulose gene as hereinbefore defined which comprises a cDNA equivalent of the *Arabidopsis thaliana rsw*1 gene. The *rsw*1 gene is a mutant cellulose gene which produces a radial root swelling phenotype as described by Baskin *et al* (1992). The present inventors have shown herein that the *rsw*1 gene also produces reduced inflorescence length, reduced fertility, misshapen epidermal cells, reduced cellulose content and the accumulation of non-crystalline β-1,4-glucan, amongst others, when expressed in plant cells. The *rsw*1 nucleotide sequence is a further variant of the nucleotide sequences set forth in SEQ ID Nos:3 and 5. The amino acid sequence set forth in SEQ ID NO:12 relates to the rsw1 polypeptide encoded by the mutant *rsw*1 gene sequence set forth in SEQ ID No:11.

15 The nucleotide sequence shown in SEQ ID NO:13 relates to a cellulose gene as hereinbefore defined which comprises a cDNA equivalent of the *Oryza sativa RSW*1 or *RSW*1-like gene. The nucleotide sequence is closely-related to the *Arabidopsis thaliana RSW*1 and *rsw*1 nucleotide sequences set forth herein (SEQ ID Nos:1, 3, 4, 5, 7, 9 and 11). The amino acid sequence set forth in SEQ ID NO:14 relates to the polypeptide encoded by the *RSW*1 or 20 *RSW*1-like gene sequences set forth in SEQ ID No:13.

Those skilled in the art will be aware of procedures for the isolation of further cellulose genes to those specifically described herein, for example further cDNA sequences and genomic gene equivalents, when provided with one or more of the nucleotide sequences set forth in SEQ 25 ID Nos:1, 3, 4, 5, 7, 9, 11, or 13. In particular, hybridisations may be performed using one or more nucleic acid hybridisation probes comprising at least 10 contiguous nucleotides and preferably at least 50 contiguous nucleotides derived from the nucleotide sequences set forth herein, to isolate cDNA clones, mRNA molecules, genomic clones from a genomic library (in particular genomic clones containing the entire 5' upstream region of the gene including 30 the promoter sequence, and the entire coding region and 3'-untranslated sequences), and/or

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synthetic oligonucleotide molecules, amongst others. The present invention clearly extends to such related sequences.

The invention further extends to any homologues, analogues or derivatives of any one or 5 more of SEQ ID Nos:1, 3, 4, 5, 7, 9, 11 or 13.

A further aspect of the present invention contemplates a nucleic acid molecule which encodes or is complementary to a nucleic acid molecule which encodes, a polypeptide which is required for cellulose biosynthesis in a plant, such as cellulose synthase, and which is capable of hybridising under at least low stringency conditions to the nucleic acid molecule set forth in any one or more of SEQ ID Nos:1, 3, 4, 5, 7, 9, 11 or 13, or to a complementary strand thereof.

As an exemplification of this embodiment, the present inventors have shown that it is possible to isolate variants of the *Arabidopsis thaliana RSW*1 gene sequence set forth in SEQ ID NO:3, by hybridization under low stringency conditions. Such variants include related sequences derived from *Gossypium hirsutum* (cotton), *Eucalyptus ssp.* and *A. thaliana*. Additional variant are clearly encompassed by the present invention.

20 Preferably, the nucleic acid molecule further comprises a nucleotide sequence which encodes, or is complementary to a nucleotide sequence which encodes, a cellulose synthase polypeptide, more preferably a cellulose synthase which is associated with the primary or secondary plant cell wall of the plant species from which said nucleic acid molecule was derived.

25

More preferably, the nucleic acid molecule according to this aspect of the invention encodes or is complementary to a nucleic acid molecule which encodes, a polypeptide which is required for cellulose biosynthesis in a plant, such as cellulose synthase, and which is capable of hybridising under at least medium stringency conditions to the nucleic acid molecule set 30 forth in any one or more of SEQ ID Nos:1, 3, 4, 5, 7, 9, 11 or 13, or to a complementary

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strand thereof.

Even more preferably, the nucleic acid molecule according to this aspect of the invention encodes or is complementary to a nucleic acid molecule which encodes, a polypeptide which 5 is required for cellulose biosynthesis in a plant, such as cellulose synthase, and which is capable of hybridising under at least high stringency conditions to the nucleic acid molecule set forth in any one or more of SEQ ID Nos:1, 3, 4, 5, 7, 9, 11 or 13, or to a complementary strand thereof.

10 For the purposes of defining the level of stringency, a low stringency is defined herein as being a hybridisation and/or a wash carried out in 6xSSC buffer, 0.1% (w/v) SDS at 28°C. Generally, the stringency is increased by reducing the concentration of SSC buffer, and/or increasing the concentration of SDS and/or increasing the temperature of the hybridisation and/or wash. A medium stringency comprises a hybridisation and/or a wash carried out in 0.2xSSC-2xSSC buffer, 0.1% (w/v) SDS at 42°C to 65°C, while a high stringency comprises a hybridisation and/or a wash carried out in 0.1xSSC-0.2xSSC buffer, 0.1% (w/v) SDS at a temperature of at least 55°C. Conditions for hybridisations and washes are well understood by one normally skilled in the art. For the purposes of further clarification only, reference to the parameters affecting hybridisation between nucleic acid molecules is found in pages 20 2.10.8 to 2.10.16. of Ausubel et al. (1987), which is herein incorporated by reference.

In an even more preferred embodiment of the invention, the isolated nucleic acid molecule further comprises a sequence of nucleotides which is at least 40% identical to at least 10 contiguous nucleotides derived from any one or more of SEQ ID Nos:1, 3, 4, 5, 7, 9, 11 or 25 13, or a complementary strand thereof.

Still more preferably, the isolated nucleic acid molecule further comprises a sequence of nucleotides which is at least 40% identical to at least 50 contiguous nucleotides derived from the sequence set forth in any one or more of SEQ ID Nos:1, 3, 4, 5, 7, 9, 11 or 13, or a 30 complementary strand thereof.

The present invention is particularly directed to a nucleic acid molecule which is capable of functioning as a cellulose gene as hereinbefore defined, for example a cellulose synthase gene such as, but not limited to, the *Arabidopsis thaliana*, *Oryza sativa*, wheat, barley, maize, *Brassica ssp.*, *Gossypium hirsutum* or *Eucalyptus ssp.* cellulose synthase genes, amongst others. The subject invention clearly contemplates additional cellulose genes to those specifically described herein which are derived from these plant species.

The invention further contemplates other sources of cellulose genes such as but not limited to, tissues and cultured cells of plant origin. Preferred plant species according to this embodiment include hemp, jute, flax and woody plants including, but not limited to *Pinus ssp.*, *Populus ssp.*, *Picea spp.*, amongst others.

A genetic sequence which encodes or is complementary to a sequence which encodes a polypeptide which is involved in cellulose biosynthesis may correspond to the naturally occurring sequence or may differ by one or more nucleotide substitutions, deletions and/or additions. Accordingly, the present invention extends to cellulose genes and any functional genes, mutants, derivatives, parts, fragments, homologues or analogues thereof or non-functional molecules but which are at least useful as, for example, genetic probes, or primer sequences in the enzymatic or chemical synthesis of said gene, or in the generation of 20 immunologically interactive recombinant molecules.

In a particularly preferred embodiment, the cellulose genetic sequences are employed to identify and isolate similar genes from plant cells, tissues, or organ types of the same species, or from the cells, tissues, or organs of another plant species.

25

According to this embodiment, there is contemplated a method for identifying a related cellulose gene or related cellulose genetic sequence, for example a cellulose synthase or cellulose synthase-like gene, said method comprising contacting genomic DNA, or mRNA, or cDNA with a hybridisation effective amount of a first cellulose genetic sequence 30 comprising any one or more of SEQ ID Nos:1, 3, 4, 5, 7, 9, 11 or 13, or a complementary

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sequence, homologue, analogue or derivative thereof derived from at least 10 contiguous nucleotides of said first sequence, and then detecting said hybridisation.

Preferably, the first genetic sequence comprises at least 50 contiguous nucleotides, even more 5 preferably at least 100 contiguous nucleotides and even more preferably at least 500 contiguous nucleotides, derived from any one or more of SEQ ID Nos:1, 3, 4, 5, 7, 9, 11 or 13, or a complementary strand, homologue, analogue or derivative thereof.

The related cellulose gene or related cellulose genetic sequence may be in a recombinant 10 form, in a virus particle, bacteriophage particle, yeast cell, animal cell, or a plant cell. Preferably, the related cellulose gene or related cellulose genetic sequence is derived from a plant species, such as a monocotyledonous plant or a dicotyledonous plant selected from the list comprising Arabidopsis thaliana, wheat, barley, maize, Brassica ssp., Gossypium hirsutum (cotton), Oryza sativa (rice), Eucalyptus ssp., hemp, jute, flax, and woody plants including, but not limited to Pinus ssp., Populus ssp., Picea spp., amongst others.

More preferably, related cellulose gene or related cellulose genetic sequence is derived from a plant which is useful in the fibre or timber industries, for example Gossypium hirsutum (cotton), hemp, jute, flax, Eucalyptus ssp. or Pinus ssp., amongst others. Alternatively, the related cellulose gene or related cellulose genetic sequence is derived from a plant which is useful in the cereal or starch industry, for example wheat, barley, rice or maize, amongst others.

In a particularly preferred embodiment, the first cellulose genetic sequence is labelled with 25 a reporter molecule capable of giving an identifiable signal (e.g. a radioisotope such as <sup>32</sup>P or <sup>35</sup>S or a biotinylated molecule).

An alternative method contemplated in the present invention involves hybridising two nucleic acid "primer molecules" to a nucleic acid "template molecule" which comprises a related 30 cellulose gene or related cellulose genetic sequence or a functional part thereof, wherein the

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first of said primers comprises contiguous nucleotides derived from any one or more of SEQ ID Nos:1, 3, 4, 5, 7, 9, 11 or 13 or a homologue, analogue or derivative thereof and the second of said primers comprises contiguous nucleotides complementary to any one or more of SEQ ID Nos:1, 3, 4, 5, 7, 9, 11 or 13. Specific nucleic acid molecule copies of the template molecule are amplified enzymatically in a polymerase chain reaction, a technique that is well known to one skilled in the art.

In a preferred embodiment, each nucleic acid primer molecule is at least 10 nucleotides in length, more preferably at least 20 nucleotides in length, even more preferably at least 30 nucleotides in length, still more preferably at least 40 nucleotides in length and even still more preferably at least 50 nucleotides in length.

Furthermore, the nucleic acid primer molecules consists of a combination of any of the nucleotides adenine, cytidine, guanine, thymidine, or inosine, or functional analogues or derivatives thereof which are at least capable of being incorporated into a polynucleotide molecule without having an inhibitory effect on the hybridisation of said primer to the template molecule in the environment in which it is used.

Furthermore, one or both of the nucleic acid primer molecules may be contained in an aqueous mixture of other nucleic acid primer molecules, for example a mixture of degenerate primer sequences which vary from each other by one or more nucleotide substitutions or deletions. Alternatively, one or both of the nucleic acid primer molecules may be in a substantially pure form.

25 The nucleic acid template molecule may be in a recombinant form, in a virus particle, bacteriophage particle, yeast cell, animal cell, or a plant cell. Preferably, the nucleic acid

template molecule is derived from a plant cell, tissue or organ, in particular a cell, tissue or organ derived from a plant selected from the list comprising Arabidopsis thaliana, Oryza sativa, wheat, barley, maize, Brassica ssp., Gossypium hirsutum and Eucalyptus ssp., hemp, jute, flax, and woody plants including, but not limited to Pinus ssp., Populus ssp., Picea 5 spp., amongst others.

Those skilled in the art will be aware that there are many known variations of the basic polymerase chain reaction procedure, which may be employed to isolate a related cellulose gene or related cellulose genetic sequence when provided with the nucleotide sequences set 10 forth in any one or more of SEQ ID Nos:1, 3, 4, 5, 7, 9, 11 or 13. Such variations are discussed, for example, in McPherson et al (1991). The present invention extends to the use of all such variations in the isolation of related cellulose genes or related cellulose genetic sequences using the nucleotide sequences embodied by the present invention.

- 15 The isolated nucleic acid molecule according to any of the further embodiments may be cloned into a plasmid or bacteriophage molecule, for example to facilitate the preparation of primer molecules or hybridisation probes or for the production of recombinant gene products. Methods for the production of such recombinant plasmids, cosmids, bacteriophage molecules or other recombinant molecules are well-known to those of ordinary skill in the art and can be accomplished without undue experimentation. Accordingly, the invention further extends to any recombinant plasmid, bacteriophage, cosmid or other recombinant molecule comprising the nucleotide sequence set forth in any one or more of SEQ ID Nos:1, 3, 4, 5, 7, 9, 11 or 13, or a complementary sequence, homologue, analogue or derivative thereof.
- 25 The nucleic acid molecule of the present invention is also useful for developing genetic constructs which express a cellulose genetic sequence, thereby providing for the increased expression of genes involved in cellulose biosynthesis in plants, selected for example from the list comprising Arabidopsis thaliana, Oryza sativa, wheat, barley, maize, Brassica ssp., Gossypium hirsutum and Eucalyptus ssp., hemp, jute, flax, and woody plants including, but 30 not limited to Pinus ssp., Populus ssp., Picea spp., amongst others. The present invention

particularly contemplates the modification of cellulose biosynthesis in cotton, hemp, jute, flax, Eucalyptus ssp. and Pinus ssp., amongst others.

The present inventors have discovered that the genetic sequences disclosed herein are capable of being used to modify the level of non-crystalline β-1,4,-glucan, in addition to altering cellulose levels when expressed, particularly when expressed in plants cells. In particular, the Arabidopsis thaliana rsw1 mutant has increased levels of non-crystalline β-1,4,-glucan, when grown at 31°C, compared to wild-type plants, grown under identical conditions. The expression of a genetic sequence described herein in the antisense orientation in transgenic plants grown at only 21°C is shown to reproduce many aspects of the rsw1 mutant phenotype.

Accordingly, the present invention clearly extends to the modification of non-crystalline β-1,4,-glucan biosynthesis in plants, selected for example from the list comprising Arabidopsis thaliana. Oryza sativa, wheat, barley, maize, Brassica ssp., Gossypium hirsutum and 15 Eucalyptus ssp., hemp, jute, flax, and woody plants including, but not limited to Pinus ssp., Populus ssp., Picea spp., amongst others. The present invention particularly contemplates the modification of non-crystalline β-1,4,-glucan biosynthesis in cotton, hemp, jute, flax, Eucalyptus ssp., and Pinus ssp., amongst others.

20 The present invention further extends to the production and use of non-crystalline  $\beta$ -1,4-glucan and to the use of the glucan to modify the properties of plant cell walls or cotton fibres or wood fibres. Such modified properties are described herein (Example 13).

The inventors have discovered that the rsw1 mutant has altered carbon partitioning compared to wild-type plants, resulting in significantly higher starch levels therein. The isolated nucleic acid molecules provided herein are further useful for altering the carbon partitioning in a cell. In particular, the present invention contemplates increased starch production in transgenic plants expressing the nucleic acid molecule of the invention in the antisense orientation or alterntively, expressing a ribozyme or co-suppression molecule comprising the nucleic acid sequence of the invention.

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The invention further contemplates reduced starch and/or non-crystalline  $\beta$ -1.4-glucan product in transgenic plants expressing the nucleic acid molecule of the invention in the sense orientation such that cellulose production is increased therein.

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5 Wherein it is desired to increase cellulose production in a plant cell, the coding region of a cellulose gene is placed operably behind a promoter, in the sense orientation, such that a cellulose gene product is capable of being expressed under the control of said promoter sequence. In a preferred embodiment, the cellulose genetic sequence is a cellulose synthase genomic sequence, cDNA molecule or protein-coding sequence.

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In a particularly preferred embodiment, the cellulose genetic sequence comprises a sequence of nucleotides substantially the same as the sequence set forth in any one or more of SEQ ID Nos:1, 3, 4, 5, 7, 9, 11 or 13 or a homologue, analogue or derivative thereof.

- Wherein it is desirable to reduce the content of cellulose or to increase the content of non-crystalline β-1,4-glucan, the nucleic acid molecule of the present invention is expressed in the antisense orientation under the control of a suitable promoter. Additionally, the nucleic acid molecule of the invention is also useful for developing ribozyme molecules, or in cosuppression of a cellulose gene. The expression of an antisense, ribozyme or co-suppression molecule comprising a cellulose gene, in a cell such as a plant cell, fungal cell, insect cell. animal cell, yeast cell or bacterial cell, may also increase the solubility, digestibility or extractability of metabolites from plant tissues or alternatively, or increase the availability of carbon as a precursor for any secondary metabolite other than cellulose (e.g. starch or sucrose). By targeting the endogenous cellulose gene, expression is diminished, reduced or otherwise lowered to a level that results in reduced deposition of cellulose in the primary or secondary cell walls of the plant cell, fungal cell, insect cell. animal cell, yeast cell or bacterial cell, and more particularly, a plant cell. Additionally, or alternatively, the content of non-crystalline β-1,4-glucan is increased in such cells.
- 30 Co-suppression is the reduction in expression of an endogenous gene that occurs when one

or more copies of said gene, or one or more copies of a substantially similar gene are introduced into the cell. The present invention also extends to the use of co-suppression to inhibit the expression of a gene which encodes a cellulose gene product, such as but not limited to cellulose synthase. Preferably, the co-suppression molecule of the present invention targets a plant mRNA molecule which encodes a cellulose synthase enzyme, for example a plant, fungus, or bacterial cellulose synthase mRNA, and more preferably a plant mRNA derived from Arabidopsis thaliana, Oryza sativa, wheat, barley, maize, Brassica ssp., Gossypium hirsutum and Eucalyptus ssp., hemp, jute, flax, or a woody plant such as Pinus ssp., Populus ssp., or Picea spp., amongst others.

10

In a particularly preferred embodiment, the gene which is targeted by a co-suppression molecule, comprises a sequence of nucleotides set forth in any one or more of SEQ ID Nos:1, 3, 4, 5, 7, 9, 11 or 13, or a complement, homologue, analogue or derivative thereof.

- 15 In the context of the present invention, an antisense molecule is an RNA molecule which is transcribed from the complementary strand of a nuclear gene to that which is normally transcribed to produce a "sense" mRNA molecule capable of being translated into a polypeptide component of the cellulose biosynthetic pathway. The antisense molecule is therefore complementary to the mRNA transcribed from a sense cellulose gene or a part thereof. Although not limiting the mode of action of the antisense molecules of the present invention to any specific mechanism, the antisense RNA molecule possesses the capacity to form a double-stranded mRNA by base pairing with the sense mRNA, which may prevent translation of the sense mRNA and subsequent synthesis of a polypeptide gene product.
- 25 Preferably, the antisense molecule of the present invention targets a plant mRNA molecule which encodes a cellulose gene product, for example cellulose synthase. Preferably, the antisense molecule of the present invention targets a plant mRNA molecule which encodes a cellulose synthase enzyme, for example a plant mRNA derived from Arabidopsis thaliana, Oryza sativa, wheat, barley, maize, Brassica ssp., Gossypium hirsutum and Eucahyptus ssp., 30 hemp, jute, flax, or a woody plant such as Pinus ssp., Populus ssp., or Picea spp., amongst

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others.

In a particularly preferred embodiment, the antisense molecule of the invention targets an mRNA molecule encoded by any one or more of SEQ ID Nos:1, 3, 4, 5, 7, 9, 11 or 13, or 5 a homologue, analogue or derivative thereof.

Ribozymes are synthetic RNA molecules which comprise a hybridising region complementary to two regions, each of at least 5 contiguous nucleotide bases in the target sense mRNA. In addition, ribozymes possess highly specific endoribonuclease activity, which autocatalytically cleaves the target sense mRNA. A complete description of the function of ribozymes is presented by Haseloff and Gerlach (1988) and contained in International Patent Application No. WO89/05852.

The present invention extends to ribozyme which target a sense mRNA encoding a cellulose gene product, thereby hybridising to said sense mRNA and cleaving it, such that it is no longer capable of being translated to synthesise a functional polypeptide product. Preferably, the ribozyme molecule of the present invention targets a plant mRNA molecule which encodes a cellulose synthase enzyme, for example a plant mRNA derived from Arabidopsis thaliana, Gossypium hirsutum (cotton), Oryza sativa (rice), Eucalyptus ssp., hemp, jute, flax, or a woody plant such as Pinus ssp., Populus ssp., or Picea spp., amongst others.

In a particularly preferred embodiment, the ribozyme molecule will target an mRNA encoded by any one or more of SEQ ID Nos:1, 3, 4, 5, 7, 9, 11 or 13, or a homologue, analogue or derivative thereof.

25

According to this embodiment, the present invention provides a ribozyme or antisense molecule comprising at least 5 contiguous nucleotide bases derived from any one or more of SEQ ID Nos:1, 3, 4, 5, 7, 9, 11 or 13, or a complementary nucleotide sequence or a homologue, analogue or derivative thereof, wherein said antisense or ribozyme molecule is 30 able to form a hydrogen-bonded complex with a sense mRNA encoding a cellulose gene

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product to reduce translation thereof.

In a preferred embodiment, the antisense or ribozyme molecule comprises at least 10 to 20 contiguous nucleotides derived from any one or more of SEQ ID Nos:1, 3, 4, 5, 7, 9, 11 or 13, or a complementary nucleotide sequence or a homologue, analogue or derivative thereof. Although the preferred antisense and/or ribozyme molecules hybridise to at least about 10 to 20 nucleotides of the target molecule, the present invention extends to molecules capable of hybridising to at least about 50-100 nucleotide bases in length, or a molecule capable of hybridising to a full-length or substantially full-length mRNA encoded by a cellulose gene, 10 such as a cellulose synthase gene.

Those skilled in the art will be aware of the necessary conditions, if any, for selecting or preparing the antisense or ribozyme molecules of the invention.

- 15 It is understood in the art that certain modifications, including nucleotide substitutions amongst others, may be made to the antisense and/or ribozyme molecules of the present invention, without destroying the efficacy of said molecules in inhibiting the expression of a gene encoding a cellulose gene product such as cellulose synthase. It is therefore within the scope of the present invention to include any nucleotide sequence variants, homologues, analogues, or fragments of the said gene encoding same, the only requirement being that said nucleotide sequence variant, when transcribed, produces an antisense and/or ribozyme molecule which is capable of hybridising to a sense mRNA molecule which encodes a cellulose gene product.
- DNA sequence to which it hybridises, thereby altering the form and/or function of the endogenous gene and the subsequent phenotype of the cell. According to this embodiment, at least a part of the DNA sequence defined by any one or more of SEQ ID Nos:1, 3, 4, 5, 7, 9, 11 or 13, or a related cellulose genetic sequence, may be introduced into target cells containing an endogenous cellulose gene, thereby replacing said endogenous cellulose gene.

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According to this embodiment, the polypeptide product of said cellulose genetic sequence possesses different catalytic activity and/or expression characteristics, producing in turn modified cellulose deposition in the target cell. In a particularly preferred embodiment of the invention, the endogenous cellulose gene of a plant is replaced with a gene which is merely capable of producing non-crystalline β-1,4-glucan polymers or alternatively which is capable of producing a modified cellulose having properties similar to synthetic fibres such as rayon, in which the β-1,4-glucan polymers are arranged in an antiparallel configuration relative to one another.

10 The present invention extends to genetic constructs designed to facilitate expression of a cellulose genetic sequence which is identical, or complementary to the sequence set forth in any one or more of SEQ ID Nos:1, 3, 4, 5, 7, 9, 11 or 13, or a functional derivative, part, homologue, or analogue thereof, or a genetic construct designed to facilitate expression of a sense molecule, an antisense molecule, ribozyme molecule, co-suppression molecule, or 15 gene targeting molecule containing said genetic sequence.

The said genetic construct of the present invention comprises the foregoing sense, antisense, or ribozyme, or co-suppression nucleic acid molecule, or gene-targeting molecule, placed operably under the control of a promoter sequence capable of regulating the expression of the said nucleic acid molecule in a prokaryotic or eukaryotic cell, preferably a plant cell. The said genetic construct optionally comprises, in addition to a promoter and sense, or antisense, or ribozyme, or co-suppression, or gene-targeting nucleic acid molecule, a terminator sequence.

25 The term "terminator" refers to a DNA sequence at the end of a transcriptional unit which signals termination of transcription. Terminators are 3'-non-translated DNA sequences containing a polyadenylation signal, which facilitates the addition of polyadenylate sequences to the 3'-end of a primary transcript. Terminators active in plant cells are known and described in the literature. They may be isolated from bacteria, fungi, viruses, animals 30 and/or plants. Examples of terminators particularly suitable for use in the genetic constructs

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of the present invention include the nopaline synthase (NOS) gene terminator of Agrobacterium tumefaciens, the terminator of the Cauliflower mosaic virus (CaMV) 35S gene, and the zein gene terminator from Zea mays.

- 5 Reference herein to a "promoter" is to be taken in its broadest context and includes the transcriptional regulatory sequences of a classical genomic gene, including the TATA box which is required for accurate transcription initiation, with or without a CCAAT box sequence and additional regulatory elements (i.e. upstream activating sequences, enhancers and silencers) which alter gene expression in response to developmental and/or external stimuli, or in a tissue-specific manner. A promoter is usually, but not necessarily, positioned upstream or 5', of a structural gene, the expression of which it regulates. Furthermore, the regulatory elements comprising a promoter are usually positioned within 2 kb of the start site of transcription of the gene.
- In the present context, the term "promoter" is also used to describe a synthetic or fusion molecule, or derivative which confers, activates or enhances expression of said sense, antisense, or ribozyme, or co-suppression nucleic acid molecule, in a plant cell. Preferred promoters may contain additional copies of one or more specific regulatory elements, to further enhance expression of a sense antisense, ribozyme or co-suppression molecule and/or to alter the spatial expression and/or temporal expression of said sense or antisense, or ribozyme, or co-suppression, or gene-targeting molecule. For example, regulatory elements which confer copper inducibility may be placed adjacent to a heterologous promoter sequence driving expression of a sense, or antisense, or ribozyme, or co-suppression, or gene-targeting molecule, thereby conferring copper inducibility on the expression of said molecule.

25

Placing a sense or ribozyme, or antisense, or co-suppression, or gene-targeting molecule under the regulatory control of a promoter sequence means positioning the said molecule such that expression is controlled by the promoter sequence. Promoters are generally positioned 5' (upstream) to the genes that they control. In the construction of heterologous promoter/structural gene combinations it is generally preferred to position the promoter at a

distance from the gene transcription start site that is approximately the same as the distance between that promoter and the gene it controls in its natural setting, i.e., the gene from which the promoter is derived. As is known in the art, some variation in this distance can be accommodated without loss of promoter function. Similarly, the preferred positioning of a regulatory sequence element with respect to a heterologous gene to be placed under its control is defined by the positioning of the element in its natural setting, i.e., the genes from which it is derived. Again, as is known in the art, some variation in this distance can also occur.

Examples of promoters suitable for use in genetic constructs of the present invention include viral, fungal, bacterial, animal and plant derived promoters capable of functioning in prokaryotic or eukaryotic cells. Preferred promoters are those capable of regulating the expression of the subject cellulose genes of the innvention in plants cells, fungal cells, insect cells, yeast cells, animal cells or bacterial cells, amongst others. Particularly preferred promoters are capable of regulating expression of the subject nucleic acid molecules in plant cells. The promoter may regulate the expression of the said molecule constitutively, or differentially with respect to the tissue in which expression occurs or, with respect to the developmental stage at which expression occurs, or in response to external stimuli such as physiological stresses, or plant pathogens, or metal ions, amongst others. Preferably, the promoter is capable of regulating expression of a sense, or ribozyme, or antisense, or co20 suppression molecule or gene targeting, in a plant cell. Examples of preferred promoters include the CaMV 35S promoter, NOS promoter, octopine synthase (OCS) promoter and the like.

In a most preferred embodiment, the promoter is capable of expression in any plant cell, such as, but not limited to a plant selected from the list comprising Arabidopsis thaliana, Oryza sativa, wheat, barley, maize, Brassica ssp., Gossypium hirsutum and Eucalyptus ssp., hemp, jute, flax, and woody plants including, but not limited to Pinus ssp., Populus ssp., Picea spp., amongst others.

30 In a particularly preferred embodiment, the promoter may be derived from a genomic clone

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encoding a cellulose gene product, in particular the promoter contained in the sequence set forth in SEQ ID NO:3 or SEQ ID NO:4. Preferably, the promoter sequence comprises nucleotide 1 to about 1900 of SEQ ID NO:3 or nucleotides 1 to about 700 of SEQ ID NO:4 or a homologue, analogue or derivative capable of hybridizing thereto under at least low 5 stringency conditions.

Optionally, the genetic construct of the present invention further comprises a terminator sequence.

10 In an exemplification of this embodiment, there is provided a binary genetic construct comprising the isolated nucleotide sequence of nucleotides set forth in SEQ ID NO:3. There is also provided a genetic construct comprising the isolated nucleotide sequence of nucleotides set forth in SEQ ID NO:1, in the antisense orientation, placed operably in connection with the CaMV 35S promoter.

15

In the present context, the term "in operable connection with" means that expression of the isolated nucleotide sequence is under the control of the promoter sequence with which it is connected, regardless of the relative physical distance of the sequences from each other or their relative orientation with respect to each other.

20

An alternative embodiment of the invention is directed to a genetic construct comprising a promoter or functional derivative, part, fragment, homologue, or analogue thereof, which is capable of directing the expression of a polypeptide early in the development of a plant cell at a stage when the cell wall is developing, such as during cell expansion or during cell division. In a particularly preferred embodiment, the promoter is contained in the sequence set forth in SEQ ID NO:3 or SEQ ID NO:4. Preferably, the promoter sequence comprises nucleotide 1 to about 1900 of SEQ ID NO:3 or nucleotides 1 to about 700 of SEQ ID NO:4 or a homologue, analogue or derivative capable of hybridizing thereto under at least low stringency conditions.

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The polypeptide may be a reporter molecule which is encoded by a gene such as the bacterial β-glucuronidase gene or chloramphenicol acetyltransferase gene or alternatively, the firefly luciferase gene. Alternatively, the polypeptide may be encoded by a gene which is capable of producing a modified cellulose in the plant cell when placed in combination with the normal complement of cellulose genes which are expressible therein, for example it may be a cellulose-like gene obtained from a bacterial or fungal source or a cellulose gene obtained from a plant source.

The genetic constructs of the present invention are particularly useful in the production of crop plants with altered cellulose content or structure. In particular, the rate of cellulose deposition may be reduced leading to a reduction in the total cellulose content of plants by transferring one or more of the antisense, ribozyme or co-suppression molecules described supra into a plant or alternatively, the same or similar end-result may be achieved by replacing an endogenous cellulose gene with an inactive or modified cellulose gene using gene-targeting approaches. The benefits to be derived from reducing cellulose content in plants are especially apparent in food and fodder crops such as, but not limited to maize, wheat, barley, rye, rice, barley, millet or sorghum, amongst others where improved digestibility of said crop is desired. The foregoing antisense, ribozyme or co-suppression molecules are also useful in producing plants with altered carbon partitioning such that increased carbon is available for growth, rather than deposited in the form of cellulose.

Alternatively, the introduction to plants of additional copies of a cellulose gene in the 'sense' orientation and under the control of a strong promoter is useful for the production of plants with increased cellulose content or more rapid rates of cellulose biosynthesis. Accordingly, such plants may exhibit a range of desired traits including, but not limited to modified strength and/or shape and/or properties of fibres, cell and plants, increased protection against chemical, physical or environmental stresses such as dehydration, heavy metals (e.g. cadmium) cold, heat or wind, increased resistance to attack by pathogens such as insects, nematodes and the like which physically penetrate the cell wall barrier during invasion/infection of the plant.

Alternatively, the production of plants with altered physical properties is made possible by the introduction thereto of altered cellulose gene(s). Such plants may produce β-1,4-glucan which is either non-crystalline or shows altered crystallinity. Such plants may also exhibit a range of desired traits including but not limited to, altered dietary fibre content, altered digestibility and degradability or producing plants with altered extractability properties.

Furthermore, genetic constructs comprising a plant cellulose gene in the 'sense' orientation may be used to complement the existing range of cellulose genes present in a plant, thereby altering the composition or timing of deposition of cellulose deposited in the cell wall of said plant. In a preferred embodiment, the cellulose gene from one plant species or a β-1,4-glucan synthase gene from a non-plant species is used to transform a plant of a different species, thereby introducing novel cellulose biosynthetic metabolism to the second-mentioned plant species.

- In a related embodiment, a recombinant fusion polypeptide may be produced containing the active site from one cellulose gene product fused to another cellulose gene product, wherein said fusion polypeptide exhibits novel catalytic properties compared to either 'parent' polypeptide from which it is derived. Such fusion polypeptides may be produced by conventional recombinant DNA techniques known to those skilled in the art, either by introducing a recombinant DNA capable of expressing the entire fusion polypeptide into said plant or alternatively, by a gene-targeting approach in which recombination at the DNA level occurs in vivo and the resultant gene is capable of expressing a recombinant fusion polypeptide.
- 25 The present invention extends to all transgenic methods and products described *supra*, including genetic constructs.

The recombinant DNA molecule carrying the sense, antisense, ribozyme or co-suppression molecule of the present invention and/or genetic construct comprising the same, may be 30 introduced into plant tissue, thereby producing a "transgenic plant", by various techniques

known to those skilled in the art. The technique used for a given plant species or specific type of plant tissue depends on the known successful techniques. Means for introducing recombinant DNA into plant tissue include, but are not limited to, transformation (Paszkowski et al., 1984), electroporation (Fromm et al., 1985), or microinjection of the 5 DNA (Crossway et al., 1986), or T-DNA-mediated transfer from Agrobacterium to the plant tissue. Representative T-DNA vector systems are described in the following references: An et al. (1985); Herrera-Estrella et al. (1983a,b); Herrera-Estrella et al. (1985). Once introduced into the plant tissue, the expression of the introduced gene may be assayed in a transient expression system, or it may be determined after selection for stable integration within the plant genome. Techniques are known for the in vitro culture of plant tissue, and in a number of cases, for regeneration into whole plants. Procedures for transferring the introduced gene from the originally transformed plant into commercially useful cultivars are known to those skilled in the art.

15 A still further aspect of the present invention extends to a transgenic plant such as a crop plant, carrying the foregoing sense, antisense, ribozyme, co-suppression, or gene-targeting molecule and/or genetic constructs comprising the same. Preferably, the transgenic plant is one or more of the following: Arabidopsis thaliana, Oryza sativa, wheat, barley, maize, Brassica ssp., Gossypium hirsutum and Eucalyptus ssp., hemp, jute, flax, Pinus ssp., 20 Populus ssp., or Picea spp. Additional species are not excluded.

The present invention further extends to the progeny of said transgenic plant.

Yet another aspect of the present invention provides for the expression of the subject genetic sequence in a suitable host (e.g. a prokaryote or eukaryote) to produce full length or non-full length recombinant cellulose gene products.

Hereinafter the term "cellulose gene product" shall be taken to refer to a recombinant product of a cellulose gene as hereinbefore defined. Accordingly, the term "cellulose gene product" includes a polypeptide product of any gene involved in the cellulose biosynthetic pathway in

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plants, such as, but not limited to a cellulose synthase gene product.

Preferably, the recombinant cellulose gene product comprises an amino acid sequence having the catalytic activity of a cellulose synthase polypeptide or a functional mutant, derivative part, fragment, or analogue thereof.

In a particularly preferred embodiment of the invention, the recombinant cellulose gene product comprises a sequence or amino acids that is at least 40% identical to any one or more of SEQ ID Nos:2, 6, 8, 10, 12 or 14, or a homologue, analogue or derivative thereof.

10

Single and three-letter abbreviations used for amino acid residues contained in the specification are provided in Table 1.

In the present context, "homologues" of an amino acid sequence refer to those polypeptides, enzymes or proteins which have a similar catalytic activity to the amino acid sequences described herein, notwithstanding any amino acid substitutions, additions or deletions thereto. A homologue may be isolated or derived from the same or another plant species as the species from which the polypeptides of the invention are derived.

20 "Analogues" encompass polypeptides of the invention notwithstanding the occurrence of any non-naturally occurring amino acid analogues therein.

"Derivatives" include modified peptides in which ligands are attached to one or more of the amino acid residues contained therein, such as carbohydrates, enzymes, proteins, polypeptides or reporter molecules such as radionuclides or fluorescent compounds. Glycosylated, fluorescent, acylated or alkylated forms of the subject peptides are particularly contemplated by the present invention. Additionally, derivatives of an amino acid sequence described herein which comprises fragments or parts of the subject amino acid sequences are within the scope of the invention, as are homopolymers or heteropolymers comprising two or more 30 copies of the subject polypeptides. Procedures for derivatizing peptides are well-known in the

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art.

TABLE 1

	Amino Acid	Three-letter	One-letter
		Abbreviation	Symbol
5	Alanine	Ala	Α
	Arginine	Arg	R
	Asparagine	Asn	N
	Aspartic acid	Asp	D
	Cysteine	Cys	С
10	D-alanine	Dal	x
	Glutamine	Gln	Q
	Glutamic acid	Glu	E
	Glycine	Gly	G
	Histidine	His	H
15	Isoleucine	Ile	1
	Leucine	Leu	L
	Lysine	Lys	K
	Methionine	Met	M
	Phenylalanine	Phe	F
20	Proline	Pro	P
	Serine	Ser	S
	Threonine	Thr	T
	Tryptophan	Trp	$\mathbf{w}$
	Tryosine	Tyr	Y
25	Valine	Val	v
	Any amino acid	Xaa	X

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Substitutions encompass amino acid alterations in which an amino acid is replaced with a different naturally-occurring or a non-conventional amino acid residue. Such substitutions may be classified as "conservative". in which an amino acid residue contained in a cellulose gene product is replaced with another naturally-occurring amino acid of similar character, for example Gly↔Ala, Val↔Ile↔Leu. Asp↔Glu, Lys↔Arg, Asn↔Gln or Phe↔Trp↔Tyr.

Substitutions encompassed by the present invention may also be "non-conservative", in which an amino acid residue which is present in a cellulose gene product described herein is substituted with an amino acid with different properties, such as a naturally-occurring amino acid from a different group (eg. substituted a charged or hydrophobic amino acid with alanine), or alternatively, in which a naturally-occurring amino acid is substituted with a non-conventional amino acid.

Non-conventional amino acids encompassed by the invention include, but are not limited to 15 those listed in Table 2.

Amino acid substitutions are typically of single residues, but may be of multiple residues, either clustered or dispersed.

20 Amino acid deletions will usually be of the order of about 1-10 amino acid residues, while insertions may be of any length. Deletions and insertions may be made to the N-terminus, the C-terminus or be internal deletions or insertions. Generally, insertions within the amino acid sequence will be smaller than amino- or carboxy-terminal fusions and of the order of 1-4 amino acid residues.

25

A homologue, analogue or derivative of a cellulose gene product as referred to herein may readily be made using peptide synthetic techniques well-known in the art, such as solid phase peptide synthesis and the like, or by recombinant DNA manipulations. Techniques for making substituent mutations at pre-determined sites using recombinant DNA technology, for 30 example by M13 mutagenesis, are also well-known. The manipulation of nucleic acid

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molecules to produce variant peptides, polypeptides or proteins which manifest as substitutions, insertions or deletions are well-known in the art.

The cellulose gene products described herein may be derivatized further by the inclusion or 5 attachment thereto of a protective group which prevents, inhibits or slows proteolytic or cellular degradative processes. Such derivatization may be useful where the half-life of the subject polypeptide is required to be extended, for ample to increase the amount of cellulose produced in a primary or secondary cell wall of a plant cell or alternatively, to increase the amount of protein produced in a bacterial or eukaryotic expression system. Examples of 10 chemical groups suitable for this purpose include, but are not limited to, any of the nonconventional amino acid residues listed in Table 2, in particular a D-stereoisomer or a methylated form of a naturally-occurring amino acid listed in Table 1. Additional chemical groups which are useful for this purpose are selected from the list comprising aryl or heterocyclic N-acyl substituents, polyalkylene oxide moieties, desulphatohirudin muteins, 15 alpha-muteins, alpha-aminophosphonic acids, water-soluble polymer groups such as polyethylene glycol attached to sugar residues using hydrazone or oxime groups, benzodiazepine dione derivatives, glycosyl groups such as beta-glycosylamine or a derivative thereof, isocyanate conjugated to a polyol functional group or polyoxyethylene polyol capped with diisocyanate, amongst others. Similarly, a cellulose gene product or a homologue, 20 analogue or derivative thereof may be cross-linked or fused to itself or to a protease inhibitor peptide, to reduce susceptibility of said molecule to proteolysis.

TABLE 2

	Non-conventional amino acid	Code	Non-conventional amino acid	Code
5				
	α-aminobutyric acid	Abu	L-N-methylalanine	Nmala
	$\alpha$ -amino- $\alpha$ -methylbutyrate	Mgabu	L-N-methylarginine	Nmarg
	aminocyclopropane-	Cpro	L-N-methylasparagine	Nmasn
	carboxylate		L-N-methylaspartic acid	Nmasp
0	aminoisobutyric acid	Aib	L-N-methylcysteine	Nmcys
	aminonorbornyl-	Norb	L-N-methylglutamine	Nmgln
	carboxylate		L-N-methylglutamic acid	Nmglu
	cyclohexylalanine	Chexa	L-N-methylhistidine	Nmhis
	cyclopentylalanine	Cpen	L-N-methylisolleucine	Nmile
5	D-alanine	Dai	L-N-methylleucine	Nmleu
	D-arginine	Darg	L-N-methyllysine	Nmlys
	D-aspartic acid	Dasp	L-N-methylmethionine	Nmmet
	D-cysteine	Dcys	L-N-methylnorleucine	Nmnle
	D-glutamine	Dgln	L-N-methylnorvaline	Nmnva
20	D-glutamic acid	Dglu	L-N-methylornithine	Nmorn
	D-histidine	Dhis	L-N-methylphenylalanine	Nmphe
	D-isoleucine	Dile	L-N-methylproline	Nmpro
	D-leucine	Dleu	L-N-methylserine	Nmser
	D-lysine	Dlys	L-N-methylthreonine	Nmthr
25	D-methionine	Dmet	L-N-methyltryptophan	Nmtrp
	D-ornithine	Dorn	L-N-methyltyrosine	Nmtyr
	D-phenylalanine	Dphe	L-N-methylvaline	Nmval
	D-proline	Dpro	L-N-methylethylglycine	Nmetg
	D-serine	Dser	L-N-methyl-t-butylglycine	Nmtbug
30	D-threonine	Dthr	L-norleucine	Nle

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	D-tryptophan	Dtrp	L-norvaline	Nva
	D-tyrosine	Dtyr	α-methyl-aminoisobutyrate	Maib
	D-valine	Dval	$\alpha$ -methyl- $\gamma$ -aminobutyrate	Mgabu
	D-α-methylalanine	Dmala	α-methylcyclohexylalanine	Mchexa
5	D-α-methylarginine	Dmarg	α-methylcylcopentylalanine	Mcpen
	D-α-methylasparagine	Dmasn	$\alpha$ -methyl- $\alpha$ -napthylalanine	Manap
	D-α-methylaspartate	Dmasp	α-methylpenicillamine	Mpen
	D-α-methylcysteine	Dmcys	N-(4-aminobutyl)glycine	Nglu
	D-α-methylglutamine	Dmgln	N-(2-aminoethyl)glycine	Naeg
10	D-α-methylhistidine	Dmhis	N-(3-aminopropyl)glycine	Norn
	D-α-methylisoleucine	Dmile	N-amino-α-methylbutyrate	Nmaabu
	D-α-methylleucine	Dmleu	α-napthylalanine	Anap
	D-α-methyllysine	Dmlys	N-benzylglycine	Nphe
	$D$ - $\alpha$ -methylmethionine	Dmmet	N-(2-carbamylethyl)glycine	Ngln
15	$D-\alpha$ -methylornithine	Dmorn	N-(carbamylmethyl)glycine	Nasn
	$D$ - $\alpha$ -methylphenylalanine	Dmphe	N-(2-carboxyethyl)glycine	Nglu
	$D$ - $\alpha$ -methylproline	Dmpro	N-(carboxymethyl)glycine	Nasp
	D-α-methylserine	Dmser	N-cyclobutylglycine	Nebut
	D-α-methylthreonine	Dmthr	N-cycloheptylglycine	Nchep
20	D-α-methyltryptophan	Dmtrp	N-cyclohexylglycine	Nchex
	D-α-methyltyrosine	Dmty	N-cyclodecylglycine	Ncdec
	D-α-methylvaline	Dmval	N-cylcododecylglycine	Ncdod
	D-N-methylalanine	Dnmala	N-cyclooctylglycine	Ncoct
	D-N-methylarginine	Dnmarg	N-cyclopropylglycine	Ncpro
25	D-N-methylasparagine	Dnmasn	N-cycloundecylglycine	Ncund
	D-N-methylaspartate	Dnmasp	N-(2,2-diphenylethyl)glycine	Nbhm
	D-N-methylcysteine	Dnmcys	N-(3,3-diphenylpropyl)glycine	Nbhe
	D-N-methylglutamine	Dnmgln	N-(3-guanidinopropyl)glycine	Narg
	D-N-methylglutamate	Dnmglu	N-(1-hydroxyethyl)glycine	Nthr
30	D-N-methylhistidine	Dnmhis	N-(hydroxyethyl))glycine	Nser

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	D-N-methylisoleucine	Dnmile	N-(imidazolylethyl))glycine	Nhis
	D-N-methylleucine	Dnmleu	N-(3-indolylyethyl)glycine	Nhtrp
	D-N-methyllysine	Dnmlys	N-methyl-γ-aminobutyrate	Nmgabu
	N-methylcyclohexylalanine	Nmchexa	D-N-methylmethionine	Dnmmet
	5 D-N-methylornithine	Dnmorn	N-methylcyclopentylalanine	Nmcpen
	N-methylglycine	Nala	D-N-methylphenylalanine	Dnmphe
	N-methylaminoisobutyrate	Nmaib	D-N-methylproline	Dnmpro
	N-(1-methylpropyl)glycine	Nile	D-N-methylserine	Dnmser
	N-(2-methylpropyl)glycine	Nleu	D-N-methylthreonine	Dnmthr
1	0 D-N-methyltryptophan	Dnmtrp	N-(1-methylethyl)glycine	Nval
	D-N-methyltyrosine	Dnmtyr	N-methyla-napthylalanine	Nmanap
	D-N-methylvaline	Dnmval	N-methylpenicillamine	Nmpen
	γ-aminobutyric acid	Gabu	N-(p-hydroxyphenyl)glycine	Nhtyr
	L-t-butylglycine	Tbug	N-(thiomethyl)glycine	Ncys
1	5 L-ethylglycine	Etg	penicillamine	Pen
	L-homophenylalanine	Hphe	L-α-methylalanine	Mala
	L-α-methylarginine	Marg	L-α-methylasparagine	Masn
	L-α-methylaspartate	Masp	L-α-methyl-t-butylglycine	Mtbug
	L-α-methylcysteine	Mcys	L-methylethylglycine	Metg
2	0 L-α-methylglutamine	Mgln	L-α-methylglutamate	Mglu
	L-α-methylhistidine	Mhis	L-α-methylhomophenylalanine	Mhphe
	L-α-methylisoleucine	Mile	N-(2-methylthioethyl)glycine	Nmet
	L-α-methylleucine	Mleu	L-a-methyllysine	Mlys
	L-α-methylmethionine	Mmet	L-α-methylnorleucine	Mnle
2	5 L-α-methylnorvaline	Mnva	L-α-methylornithine	Morn
	L-α-methylphenylalanine	Mphe	L-a-methylproline	Mpro
	L-a-methylserine	Mser	L-α-methylthreonine	Mthr
	L-α-methyltryptophan	Mtrp	L-\a-methyltyrosine	Mtyr
	L-α-methylvaline	Mval	L-N-methylhomophenylalanine	Nmhphe

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N-(N-(2,2-diphenylethyl) Nnbhm N-(N-(3,3-diphenylpropyl) Nnbhe carbamylmethyl)glycine carbamylmethyl)glycine
1-carboxy-1-(2,2-diphenyl- Nmbc ethylamino)cyclopropane

In an alternative embodiment of the invention, the recombinant cellulose gene product is characterised by at least one functional  $\beta$ -glycosyl transferase domain contained therein.

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The term "β-glycosyl transferase domain" as used herein refers to a sequence of amino acids which is highly conserved in different processive enzymes belonging to the class of glycosyl transferase enzymes (Saxena et al., 1995), for example the bacterial β-1,4-glycosyl transferase enzymes and plant cellulose synthase enzymes amongst others, wherein said domain possesses a putative function in contributing to or maintaining the overall catalytic activity, substrate specificity or substrate binding of an enzyme in said enzyme class. The β-glycosyl transferase domain is recognisable by the occurrence of certain amino acid residues at particular locations in a polypeptide sequence, however there is no stretch of contiguous amino acid residues comprised therein.

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As a consequence of the lack of contiguity in a β-glycosyl transferase domain, it is not a straightforward matter to isolate a cellulose gene by taking advantage of the presence of a β-glycosyl transferase domain in the polypeptide encoded by said gene. For example, the β-glycosyl transferase domain would not be easily utilisable as a probe to facilitate the rapid isolation of all β-glycosyl transferase genetic sequences from a particular organism and then to isolate from those genetic sequences a cellulose gene such as cellulose synthase.

In a preferred embodiment, the present invention provides an isolated polypeptide which: (i)contains at least one structural  $\beta$ -glycosyl transferase domain as hereinbefore defined; and

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- (ii) has at least 40% amino acid sequence similarity to at least 20 contiguous amino acid residues set forth in any one or more of SEQ ID Nos:2, 6, 8, 10, 12 or 14, or a homologue, analogue or derivative thereof.
- 5 More preferably, the polypeptide of the invention is at least 40% identical to at least 50 contiguous amino acid residues, even more preferably at least 100 amino acid residues of any one or more of SEQ ID Nos:2, 6, 8, 10, 12 or 14, or a homologue, analogue or derivative thereof.
- In a particularly preferred embodiment, the percentage similarity to any one or more of SEQ ID Nos:2, 6, 8, 10, 12 or 14 is at least 50-60%, more preferably at least 65-70%, even more preferably at least 75-80% and even more preferably at least 85-90%, including about 91% or 95%.
- 15 In a related embodiment, the present invention provides a "sequencably pure" form of the amino acid sequence described herein. "Sequencably pure" is hereinbefore described as substantially homogeneous to facilitate amino acid determination.
- In a further related embodiment, the present invention provides a "substantially homogeneous" form of the subject amino acid sequence, wherein the term "substantially homogeneous" is hereinbefore defined as being in a form suitable for interaction with an immunologically interactive molecule. Preferably, the polypeptide is at least 20% homogeneous, more preferably at least 50% homogeneous, still more preferably at least 75% homogeneous and yet still more preferably at least about 95-100% homogeneous, in 25 terms of activity per microgram of total protein in the protein preparation.

The present invention further extends to a synthetic peptide of at least 5 amino acid residues in length derived from or comprising a part of the amino acid sequence set forth in any one or more of SEQ ID Nos:2, 6, 8, 10, 12 or 14, or having at least 40% similarity thereto.

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Those skilled in the art will be aware that such synthetic peptides may be useful in the production of immunologically interactive molecules for the preparation of antibodies or as the peptide component of an immunoassay.

5 The invention further extends to an antibody molecule such as a polyclonal or monoclonal antibody or an immunologically interactive part or fragment thereof which is capable of binding to a cellulose gene product according to any of the foregoing embodiments.

The term "antibody" as used herein, is intended to include fragments thereof which are also specifically reactive with a polypeptide of the invention. Antibodies can be fragmented using conventional techniques and the fragments screened for utility in the same manner as for whole antibodies. For example, F(ab')2 fragments can be generated by treating antibody with pepsin. The resulting F(ab')2 fragment can be treated to reduce disulfide bridges to produce Fab' fragments.

15

Those skilled in the art will be aware of how to produce antibody molecules when provided with the cellulose gene product of the present invention. For example, by using a polypeptide of the present invention polyclonal antisera or monoclonal antibodies can be made using standard methods. A mammal, (e.g., a mouse, hamster, or rabbit) can be immunized with an immunogenic form of the polypeptide which elicits an antibody response in the mammal. Techniques for conferring immunogenicity on a polypeptide include conjugation to carriers or other techniques well known in the art. For example, the polypeptide can be administered in the presence of adjuvant. The progress of immunization can be monitored by detection of antibody titers in plasma or serum. Standard ELISA or other immunoassay can be used with the immunogen as antigen to assess the levels of antibodies. Following immunization, antisera can be obtained and, if desired IgG molecules corresponding to the polyclonal antibodies may be isolated from the sera.

To produce monoclonal antibodies, antibody producing cells (lymphocytes) can be harvested 30 from an immunized animal and fused with myeloma cells by standard somatic cell fusion

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procedures thus immortalizing these cells and yielding hybridoma cells. Such techniques are well known in the art. For example, the hybridoma technique originally developed by Kohler and Milstein (1975) as well as other techniques such as the human B-cell hybridoma technique (Kozbor et al., 1983), the EBV-hybridoma technique to produce human monoclonal antibodies (Cole et al., 1985), and screening of combinatorial antibody libraries (Huse et al., 1989). Hybridoma cells can be screened immunochemically for production of antibodies which are specifically reactive with the polypeptide and monoclonal antibodies isolated.

As with all immunogenic compositions for eliciting antibodies, the immunogenically effective amounts of the polypeptides of the invention must be determined empirically. Factors to be considered include the immunogenicity of the native polypeptide, whether or not the polypeptide will be complexed with or covalently attached to an adjuvant or carrier protein or other carrier and route of administration for the composition, i.e. intravenous, intramuscular, subcutaneous, etc., and the number of immunizing doses to be administered. Such factors are known in the vaccine art and it is well within the skill of immunologists to make such determinations without undue experimentation.

It is within the scope of this invention to include any second antibodies (monoclonal, polyclonal or fragments of antibodies) directed to the first mentioned antibodies discussed above. Both the first and second antibodies may be used in detection assays or a first antibody may be used with a commercially available anti-immunoglobulin antibody.

The present invention is further described by reference to the following non-limiting Figures and Examples.

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In the Figures:

Figure 1 is a photographic representation showing the inflorescence length of wild-type Arabidopsis thaliana Columbia plants (plants 1 and 3) and rsw1 plants (plants 2 and 4) grown at 21°C (plants 1 and 2) or 31°C. Plants were grown initially at 21°C until bolting commenced, the bolts were removed and the re-growth followed in plants grown at each temperature.

Figure 2 is a photographic representation of a cryo-scanning electron micrograph showing 10 misshapen epidermal cells in the cotyledons and hypocotyl of the *rsw*1 mutant when grown at 31°C for 10 days.

Figure 3 is a graphical representation of a gas chromatograph of alditol acetates of methylated sugars from a cellulose standard (top panel) and from the neutral glucan derived from shoots of rsw1 plants grown at 31°C (lower panel). The co-incident peaks show that the rsw1 glucan is 1,4-linked.

Figure 4 is a schematic representation of the contiguous region of Arabidopsis thaliana chromosome 4 (stippled box) between the cosmid markers g8300 and 06455, showing the location of overlapping YAC clones (open boxes) within the contiguous region. The position of the RSW1 locus is also indicated, approximately 1.2cM from g8300 and 0.9cM from 06455. The scale indicates 100kb in length. L, left-end of YAC; R, right-end of YAC. Above the representation of chromosome 4, the YAC fragments and cosmid clone fragments used to construct the contiguous region are indicated, using a prefix designation 25 corresponding to the YAC or cosmid from which the fragments were obtained (eg yUP9E3, yUP20B12, etc) and a suffix designation indicating whether the fragment corresponds to the right-end (RE) or left-end (LE) of the YAC clone; N, North; S, South; CAPS, cleaved amplified polymorphic sequence (Konieczny and Ausubel, 1993) version of the g8300 marker.

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Figure 5 is a schematic representation of a restriction map of construct 23H12 between the left T-DNA border (LB) and right T-DNA border (RB) sequences (top solid line), showing the position of the Arabidopsis thaliana RSW1 locus (stippled box). The line at the top of the figure indicates the region of 23H12 which is contained in construct pRSW1. The structure of the RSW1 gene between the translation start (ATG) and translation stop (TAG) codons is indicated at the bottom of the figure. Exons are indicated by filled boxes; introns are indicated by the solid black line. The alignment of EST clone T20782 to the 3'-end of the RSW1 gene, from near the end of exon 7 to the end of exon 14, is also indicated at the bottom of the figure. Restriction sites within 23H12 are as follows: B, BamHI; E, EcoRI; 10 H, HindIII; S, SalI; Sm, SmaI.

- Figure 6 is a photographic representation showing complementation of the radial root swelling phenotype of the rsw1 mutant by transformation with construct 23H12. The rsw1 mutant was transformed with 23H12 as described in Example 6. Transformed rsw1 plants (centre group of three seedlings), untransformed rsw1 plants (left group of three seedlings) and untransformed A.thaliana Columbia plants (right group of three seedlings) were grown at 21°C for 5 days and then transferred to 31°C for a further 2 days, after which time the degree of root elongation and radial root swelling was determined.
- 20 Figure 7 is a photographic representation comparing wild-type Arabidopsis thaliana Columbia plants (right-hand side of the ruler) and A.thaliana Columbia plants transformed with the antisense RSW1 construct (i.e. EST T20782 expressed in the antisense orientation under control of the CaMV 35S promoter sequence; left-hand side of the ruler), showing inflorescence shortening at 21°C in plants transformed with the antisense RSW1 construct compared to untransformed Columbia plants. The phenotype of the antisense plants at 21°C is similar to the phenotype of the rsw1 mutant at 31°C. Inflorescence height is indicated in millimetres.
- Figure 8 is a schematic representation showing the first 90 amino acid residues of 30 Arabidopsis thaliana RSW1 aligned to the amino acid sequences of homologous polypeptides

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from A. thaliana and other plant species. The shaded region indicates highly conserved sequences. Ath-A and Ath-B are closely related Arabidopsis thaliana cDNA clones identified by hybridisation screening using part of the RSW1 cDNA as a probe. S0542, rice EST clone (MAFF DNA bank, Japan); celA1 and celA2, cotton cDNA sequences expressed in cotton fibre (Pear et al, 1996); SOYSTF1A and SOYSTF1B, putative soybean bZIP transcription factors. Amino acid designations are as indicated in Table 1 incorporated herein. Conserved cysteine residues are indicated by the asterisk.

Figure 9 is a schematic representation showing the alignment of the complete amino acid sequence of Arabidopsis thaliana RSW1 to the amino acid sequences of homologous polypeptides from A. thaliana and other plant species. The shaded region indicates highly conserved sequences. Ath-A and Ath-B are closely related Arabidopsis thaliana cDNA clones identified by hybridisation screening using part of the RSW1 cDNA as a probe. S0542, rice EST clone (MAFF DNA bank, Japan); celA1, cotton genetic sequence (Pear et al, 1996); D48636, a partial cDNA clone obtained from rice (Pear et al, 1996). Amino acid designations are as indicated in Table 1 incorporated herein. Numbering indicates the amino acid position in the RSW1 sequence.

Figure 10 is a schematic representation of the RSW1 polypeptide, showing the positions of putative transmembrane helices (hatched boxes), cysteine-rich region (Cys) and aspartate residues (D) and the QVLRW signature which are conserved between RSW1 and related amino acid sequences. Regions of RSW1 which are highly-conserved between putative cellulose biosynthesis polypeptides are indicated by the dark-shaded boxes, while less-conserved regions are indicated by the light-shaded boxes.

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Figure 11 is a photographic representation of a Southern blot hybridisation of the 5'- end of the *Arabidopsis thaliana* RSW1 cDNA to *BgI*II-digested DNA derived from *A. thaliana* (lane 1) and cotton (lane 2). Hybridisations were carried out under low stringency conditions at 55°C. Arrows indicate the positions of hybridising bands.

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#### **EXAMPLE 1**

### CHARACTERISATION OF THE CELLULOSE-DEFICIENT

#### Arabidopsis thaliana MUTANT rsw1

#### 5 1. Morphology

The Arabidopsis thaliana rsw1 mutant was produced in a genetic background comprising the ecotype Columbia.

The altered root cell-shape and temperature sensitivity of the root morphology of the 10 Arabidopsis thaliana mutant rswl are disclosed, among other morphological mutants, by Baskin et al. (1992).

As shown in Figure 1, the present inventors have shown that the rsw1 mutant exhibits the surprising phenotype of having reduced inflorescence height when grown at 31°C, compared to wild-type Columbia plants grown under similar conditions. In contrast, when grown at 21°C, the inflorescence height of rsw1 is not significantly different from wild type plants grown under similar conditions, indicating that the shoot phenotype of rsw1 is conditional and temperature-dependent.

- 20 Furthermore, cryo-scanning electron microscopy of the epidermal cells of the *rsw1* mutant indicates significant abnormality in cell shape, particularly in respect of those epidermal cells forming the leaves, hypocotyl and cotyledons, when the seedlings are grown at 31°C (Figure 2).
- 25 Rosettes (terminal complexes) are the putative hexameric cellulose synthase complexes of higher plant plasma membranes (Herth, 1985). Freeze-fractured root cells of Arabidopsis thaliana rsw1 plants grown at 18°C show cellulose microfibrils and rosettes on the PF face of the plasma membrane that resembles those of wild-type A. thaliana and other angiosperms. Transferring the rsw1 mutant to 31°C reduces the number of rosettes in the 30 mutant within 30 min, leading to extensive loss after 3 hours. Plasma membrane particles

align in rows on prolonged exposure to the restrictive temperature. In contrast, there is no change in the appearance of cortical microtubules that align cellulose microfibrils, or of Golgi bodies that synthesise other wall polysaccharides and assemble rosettes.

#### 5 2. Carbohydrate content

The effect of mutations in the RSW1 gene on the synthesis of cellulose and other carbohydrates was assessed by measuring in vivo incorporation of <sup>14</sup>C (supplied as uniformly labelled glucose) into various cell wall fractions. Wild type (RSW1) and homozygous mutant rsw1 seed were germinated at 21°C on agar containing Hoagland's nutrients and 1% (w/v) 10 unlabelled glucose. After 5 d, half of the seedlings were transferred to 31°C for 1 d while the remainder were maintained at 21°C for the same time. Seedlings were covered with a solution containing Hoagland's nutrients and <sup>14</sup>C-glucose and incubated for a further 3 h at the same temperature. Rinsed roots and shoots were separated and frozen in liquid nitrogen. Tissue was homogenised in cold, 0.5 M potassium phosphate buffer (0.5M KH<sub>2</sub>PO<sub>4</sub>, pH7.0) 15 and a crude cell wall fraction collected by centrifugation at 2800 rpm. The wall fraction was extracted with chloroform/methanol [1:1 (v/v)] at 40°C for 1 hour, followed by a brief incubation at 150°C, to remove lipids. The pellet was washed successively with 2ml methanol, 2ml acetone and twice with 2ml of deionised water. Finally, the pellet was extracted successively with dimethyl sulphoxide under nitrogen to remove starch: 0.5% 20 ammonium oxalate to remove pectins; 0.1 M KOH and 3 mg/ml NaBH4 and then with 4 M KOH and 3 mg/ml NaBH<sub>4</sub> to extract hemicelluloses; boiling acetic acid/nitric acid/water [8:1:2 (v/v)], to extract any residual non-cellulosic carbohydrates and leave crystalline cellulose as the final insoluble pellet (Updegraph, 1969). All fractions were analysed by liquid scintillation counting and the counts in each fraction from the mutant were expressed 25 as a percentage of the counts in the wild type under the same conditions.

As shown in Table 3, mutant and wild type plants behave in quite similar fashion at 21°C (the permissive temperature) whereas, at the restrictive temperature of 31°C, the incorporation of <sup>14</sup>C into cellulose is severely inhibited (to 36% of wild type) by the rsw1 mutation. The data in Table 3 indicate that cellulose synthesis is specifically inhibited in

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the rsw1 mutant. The wild type RSW1 gene is therefore involved quite directly in cellulose synthesis and changing its sequence by mutation changes the rate of synthesis.

5 TABLE 3

Counts in fractions from rswl plants expressed as a % of counts in comparable fraction from wild type plants					
Pectins		Hemicelluloses		Cellulose	
21°C	31°C	21°C	31°C	21°C	31°C
125	104	111	101	80	36

10

In homozygous mutant rsw1 plants, the pectin fraction extracted by ammonium oxalate contained abundant glucose, atypical of true uronic acid-rich pectins. The great majority of the glucose remained in the supernatant when cetyltrimethylammonium bromide precipitated 15 the negatively charged pectins.

#### 3. Non-crystalline β-1,4-glucan content

The quantity of cellulose and the quantity of a non-crystalline β-1,4-glucan recovered from the ammonium oxalate fraction were determined for seedlings of wild type Columbia and for 20 backcrossed, homozygous rswl that were grown for either 7 days at 21°C or alternatively, for 2 days at 21°C and 5 days at 31°C, on vertical agar plates containing growth medium (Baskin et al., 1992) plus 1% (w/v) glucose, and under continuous light (90 μmol m<sup>-2</sup> s<sup>-1</sup>). Roots and shoots were separated from about 150 seedlings, freeze-dried to constant weight and ground in a mortar and pestle with 3 ml of cold 0.5 M potassium phosphate buffer (pH 7.0). The 25 combined homogenate after two buffer rinses (2ml each) was centrifuged at 2800 x g for 10 min. After washing the pellet fraction twice with 2 ml buffer and twice with 2 ml distilled water, the pellet, comprising the crude cell wall fraction, and the pooled supernatants, comprising the phosphate buffer fraction were retained. The crude cell wall pellet fraction was stirred with two 3 ml aliquots of chloroform/methanol [1:1 (v/v)] for 1 hour at 40°C, 2 ml of methanol at 40°C for 30 min, 2 ml of acetone for 30 min, and twice with water. The whole

procedure repeated in the case of shoots. Combined supernatants were dried in a nitrogen stream. The pellet was successively extracted with: (i)3 ml of DMSO- water 9:1 [v/v], sealed under nitrogen, overnight with shaking, followed by two 2ml extractions using DMSO/water and three 2ml water washes; (ii) 3ml of ammonium oxalate (0.5 %) at 100°C for 1 hour, followed by two water washes; (iii) 3ml of 0.1 M KOH containing 1mg/ ml sodium borohydride, for 1 hour at 25°C (repeated once for root material or twice for shoot material), with a final wash with 2 ml water; (iv) 3 ml of 4 M KOH containing 1 mg/ml sodium borohydride, for 1 hour at 25°C (repeated once for root material or twice for shoot material). The final pellet was boiled with intermittent stirring in 3 ml of acetic acid-nitric acid-water [8:1:2 (v/v)] (Updegraph 1969), combined with 2 water washes, and diluted with 5 ml water.

The insoluble residue of cellulose was solubilised in 67% (v/v) H<sub>2</sub>SO<sub>4</sub>, shown to contain greater than 97% (w/v) glucose using GC/MS (Fisons AS800/MD800) of alditol acetates (Doares *et al.*, 1991) and quantified in three independent samples by anthrone/H<sub>2</sub>SO<sub>4</sub> reaction.

15 Results of GC/MS for pooled replica samples are presented in Table 4.

The non-crystalline β-1,4-glucan was recovered as the supernatant from the ammonium oxalate fraction when anionic pectins were precipitated by overnight incubation at 37 °C with 2% (w/v) cetyltrimethylammonium bromide (CTAB) and collected by centrifugation at 2800 x g for 10 min. The glucan (250 µg/ml) or starch (Sigma; 200 µg/ml) were digested with mixtures of endocellulase (EC 3.2.1.4; Megazyme, Australia) from *Trichoderma* and almond β-glucosidase (EC 3.2.1.21; Sigma), or *Bacillus sp.* α-amylase (EC 3.2.1.1; Sigma) and rice α-glucosidase (EC 3.2.1.20; Sigma).

- 25 The material recovered in the supernatant from the ammonium oxalate fraction was shown to contain a pure β-1,4-glucan by demonstrating that: (i) only glucose was detectable when it was hydrolysed by 2 M TFA in a sealed tube for 1 h at 120°C in an autoclave, the supernatant (2000 g for 5 min) was dried under vacuum at 45°C to remove TFA and glucose was determined by GC/MS;
  (ii) methylation (Needs and
- 30 Selvendran 1993) gave a dominant peak resolved by thin layer chromatography and by GC/MS

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that was identical to that from a cellulose standard and so indicative of 1,4-linked glucan (Figure 3); and

(iii) the endo-cellulase and β-1,4-glucosidase mixture released 83 % of the TFA-releasable glucose from the glucan produced by rswl at 31°C while 5 the α-amylase/α-glucosidase mixture released no glucose from the glucan. Conversely, the α-amylase/α-glucosidase mixture released 95% of the TFA-releasable glucose from a starch sample, while the endo-cellulase/β-1,4-glucosidase mixture released no glucose from starch.

Extractability of the glucan using ammonium oxalate, and the susceptibility of the glucan to endocellulase/β-glucosidase and TFA hydrolysis indicate that the glucan in the rswl mutant is not crystalline, because it is the crystallinity of glucan which makes cellulose resistant to extraction and degradation.

Table 4 shows the quantity of glucose in cellulose determined by the anthrone/H<sub>2</sub>SO<sub>4</sub> reaction and the quantity in the non-crystalline glucan after TFA hydrolysis, for shoots of wild type and mutant rsw1 Arabidopsis plants. The data indicate that the production of cellulose and of the non-crystalline β-1,4-glucan can be manipulated by mutational changes in the RSW1 gene.

TABLE 4

Glucose contents of cellulose and of the ammonium oxalate-extractable glucan

	wild type		rs	wl
	21°C	31°C	21°C	31°C
Cellulose	273+28	363+18*	218+20	159+19*
Glucan	22	58	24	195

All values nmol glucose mg-1 plant dry weight + sd (n=3).

25 \* Differences significant at 0.001 % level.

#### 4. Starch content

The quantity of starch recovered in the DMSO fraction from roots in the experiment described above was also determined by the anthrone/H<sub>2</sub>SO<sub>4</sub> extraction (Table 5).

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As shown in Table 5, the level of starch deposited in the rsw1 mutant is 4-fold that detectable in the roots of wild-type plants at the restrictive temperature of 31°C. A similar rise in starch is also seen if the data are expressed as nmol glucose per plant. There is no detectable difference in deposition at starch between rsw1 plants and wild-type plants at 5 21°C.

TABLE 5

Quantity of starch (nmol glucose per mg dry weight of seedling) extracted from roots of rsw1 and wild type seedlings

ſ		Phenotype		
10	Temperature	Wild-type	rsw1 mutant	
	21°C	22	18	
	31°C	37	126	

The composition of cell walls in the rsw1 mutant plant compared to wild type plants at the 15 restrictive temperature of 31°C, is summarised in Table 6.

TABLE 6

Mol% composition of cell walls from shoots of rsw1 and wild-type seedlings grown at 31°C

	Phenotype		
Cell wall component	Wild-type	rsw1 mutant	
Crystalline cellulose	38.4	16.5	
Non- crystalline β-1,4-glucan	8.5	27.1	
Pectin	37.1	36.3	
Alkali-soluble	15.6	19.8	
Acid-soluble	0.3	0.4	

25

20

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In conclusion, the rsw1 mutation disassembles cellulose synthase complexes in the plasma membrane, reduces cellulose accumulation and causes  $\beta-1,4$ -glucan to accumulate in a non-crystalline form.

5

# EXAMPLE 2 MAPPING OF YAC CLONES TO THE rsw1 LOCUS

The rsw1 locus in the mutant Arabidopsis thaliana plant described in Example 1 above was 10 mapped to chromosome 4 of A. thaliana using RFLP gene mapping techniques(Chang et al., 1988: Nam et al., 1989) to analyse the F<sub>2</sub> or F<sub>3</sub> progeny derived from a Columbia (Co)/Landsberg (Ler) cross. In particular, the rsw1 mutation was shown to be linked genetically to the ga5 locus, which is a chromosome 4 visual marker in A. thaliana.

15 Based on an analysis of map distances and chromosomal break points in 293 F<sub>2</sub> or F<sub>3</sub> progeny derived from a Columbia (Co)/Landsberg (Ler) cross, rswl was localised to an approximately 2.1 cM region between the RFLP markers g8300 and 06455, approximately 1.2cM south of the CAPS (cleaved amplified polymorphic sequence; Konieczny and Ausubel, 1993) version of the g8300 marker (Figure 4).

20

The interval between g8300 and 06455 in which rsw1 residues was found to be spanned by an overlapping set of Yeast Artificial Chromosome (YAC) clones. The clones were obtained from Plant Industry, Commonwealth Scientific and Industrial Research Organisation, Canberra, Australia. The YACs were positioned in the g8300/06455 interval by hybridisation using known DNA molecular markers (from within the interval) and DNA fragments from the ends of the YACs. The length of the interval was estimated to comprise 900kb of DNA.

Refined gene mapping of recombinants within the region spanned by YAC clones established 30 the genetic distance between the RFLP marker g8300 and the rsw1 locus.

- 50 -

The combination of genetic map distance data and the mapping of YAC clones within the region further localised the rswl locus to the YAC clone designated yUP5C8.

#### 5 EXAMPLE 3

### MAPPING OF cDNA CLONES TO THE YAC CLONE YUP5C8

An Arabidopsis thaliana cDNA clone designated T20782 was obtained from the public Arabidopsis Resource Centre, Ohio State University, 1735 Neil Avenue, Columbus, OH 43210, United States of America. The T20782 cDNA clone was localised broadly to the DNA interval on Arabidopsis chromosome 4 between the two markers g8300 and 06455 shown in Figure 4. Using a polymerase chain reaction (PCR) based approach DNA primers (5'-AGAACAGCAGATACACGGA-3' and 5'-CTGAAGAAGGCTGGACAAT-3') designed to the T20782 cDNA nucleotide sequence were used to screen Arabidopsis YAC clone libraries. The T20782 cDNA clone was found to localise to YACs (CIC1F9, CIC10E9, CIC11D9) identified on the Arabidopsis chromosome 4 g8300 and 06455 interval (Figure 4). The same approach was used to further localise clone T20782 to YAC clone yUP5C8, the same YAC designated to contain the rsw1 locus in the same chromosome interval (Figure 4).

20

Furthermore, amplification of the YAC clone yUP5C8 using primers derived from T20782 produces a 500bp fragment containing two putative exons identical to part of the T20782 nucleotide sequence, in addition to two intron sequences.

25 The cDNA T20782 was considered as a candidate gene involved in cellulose biosynthesis.

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#### **EXAMPLE 4**

### **NUCLEOTIDE SEQUENCE ANALYSIS OF THE CDNA CLONE T20782**

5 The nucleotide sequence of the cDNA clone T20782 is presented in SEQ ID NO: 1. The nucleotide sequence was obtained using a Dye Terminator Cycle Sequencing kit (Perkin Elmer cat. #401384) as recommended by the manufacturer. Four template clones were used for nucleotide sequencing to generate the sequence listed. The first template was the cDNA clone T20782. This template was sequenced using the following sequencing primers:

10

- a)5'-CAATGCATTCATAGCTCCAGCCT-3'
- b)5'-AAAAGGCTGGAGCTATGAATGCAT-3'
- c)5'-TCACCGACAGATTCATCATACCCG-3'
- d)5'- GACATGGAATCACCTTAACTGCC-3'
- 15 e)5'-CCATTCAGTCTTGTCTTCGTAACC-3'
  - **1)5'-GGTTACGAAGACAAGACTGAAATGG-3'**
  - g)5'-GAACCTCATAGGCATTGTGGGCTGG-3'
  - h)5'-GCAGGCTCTATATGGGTATGATCC-3'
  - i)Standard M13 forward sequencing primer.
- j)Standard T7 sequencing primer.

The second template clone (T20782 SphI deletion clone) was constructed by creating a DNA deletion within the T20782 clone. The T20782 clone was digested with the restriction enzyme SphI, the enzyme was heat-killed, the DNA ligated and electroporated into NM522 E. coli host cells. The T20782 SphI deletion clone was then sequenced using a standard M13 forward sequencing primer. Two other deletion clones were made for DNA sequencing in a similar fashion but the restriction enzymes EcoRI and SmaI were used. The T20782 EcoRI deletion clone and the T20782 SmaI deletion clone were sequenced using a standard T7 sequencing primer. The DNA sequence shown in SEQ ID NO:1 is for one DNA strand only however those skilled in the art will be able to generate the nucleotide sequence of the

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complementary strand from the data provided.

The amino acid sequence encoded by clone T20782 was derived and is set forth in SEQ ID NO:2.

5

The T20782 clone encodes all but the first Aspartate (D) residue of the D, D, D, QXXRW signature conserved in the general architecture of β-glycosyl transferases. In particular, T20782 encodes 5 amino acid residues of the D, D, D, QXXRW signature, between amino acid positions 109 and 370 of SEQ ID NO:2. The conserved Aspartate, Aspartate,

- 10 Glutamine. Arginine and Tryptophan amino acid residues are shown below, in bold type, with the local amino acid residues also indicated:
  - 1. Amino acid residues 105 to 113 of SEQ ID NO:2:

#### LLNVDCDHY;

15 2. Amino acid residues 324 to 332 of SEQ ID NO:2:

SVTEDILTG; and

3. Amino acid residues 362 to 374 of SEQ ID NO:2:

#### DRLNOVLRWALGS.

- 20 It must be noted that these invariable amino acids merely indicate that the T20782 derived amino acid sequence belongs to a very broad group of glycosyl transferases. Some of these enzymes such as cellulose synthase, chitin synthase, alginate synthase and hyaluronic acid synthase produce functionally very different compounds.
- 25 The presence of the conserved amino acid residues merely indicate that the T20782 clone may encode a β-glycosyl transferase protein such as the cellulose gene product, cellulose synthase. The fact that the clone localises in the vicinity of a gene involved in cellulose biosynthesis is the key feature which now focus interest on the T20782 clone as a candidate for the RSW1 (cellulose synthase) gene.

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The T20782 potentially codes for a cellulose synthase.

#### **EXAMPLE 5**

### 5 NUCLEOTIDE SEQUENCE ANALYSIS OF THE GENOMIC CLONE 23H12

Clone 23H12 contains approximately 21kb of Arabidopsis thaliana genomic DNA in the region between the left border and right border T-DNA sequences, and localises to the RSW1 candidate YAC yUP5C8. Clone 23H12 was isolated by hybridisation using EST20782 insert DNA, from a genomic DNA library made for plant transformation. Cosmid 12C4 was also shown to hybridize to the cDNA clone T20782, however this cosmid appears to comprise a partial genomic sequence corresponding to the related Ath-A cDNA sequence set forth in SEQ ID NO:7, for which the corresponding amino acid sequence is set forth in SEQ ID NO:8.

15

A restriction enzyme map of clone 23H12 is presented in Figure 5.

Nucleotide sequence of 8411bp of genomic DNA in the binary cosmid clone 23H12 was obtained (SEQ ID NO:3) by primer walking along the 23H12 template, using a Dye 20 Terminator Cycle Sequencing kit (Perkin Elmer cat. #401384) as recommended by the manufacturer. The following primers at least, were used for DNA sequencing of the 23H12 clone DNA:

	a)cs1-R	5'-CAATGCATTCATAGCTCCAGCCT-3'
25	b)cs1-F	5'-AAAAGGCTGGAGCTATGAATGCAT-3'
	c)up	5'-AGAACAGCAGATACACGGA-3'
	d)ve76-R2	5'-ATCCGTGTATCTGCTGTTCTTACC-3'
	e)est1-R	5'-AATGCTCTTGTTGCCAAAGCAC-3'
	f)sve76-F	5'-ATTGTCCAGCCTTCTTCAGG-3'
30	g)ve76-R	5'-CTGAAGAAGGCTGGACAATGC-3'

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h)B12-R1	5'-AGGTAAGCATAGCTGAACCATC-3'
i)B12-R2	5'-AGTAGATTGCAGATGGTTTTCTAC-3'
j)B12-R3	5'-TTCAATGGGTCCACTGTACTAAC-3'
k)B12-R4	5'-ATTCAGATGCACCATTGTC-3'

5

The structure of the RSW1 gene contained in cosmid clone 23H12 is also presented in Figure 5. As shown therein, coding sequences in 23H12, from the last 12 bp of exon 7 to the end of exon 14, correspond to the full T20782 cDNA sequence (i.e. SEQ ID NO:1). The nucleotide sequences of the RSW1 gene comprising exons 1 to 8 were amplified from 10 A.thaliana Columbia double-stranded cDNA, using amplification primers upstream of the RSW1 start site and a primer internal to the EST clone T20782.

The exons in the RSW1 gene range from 81bp to 585bp in length and all 5' and 3' intron/exon splice junctions conform to the conserved intron rule.

15

The RSW1 transcript comprises a 5'-untranslated sequence of at least 70bp in length, a 3243bp coding region and a 360bp 3'-untranslated region. Northern hybridization analyses indicate that the RSW1 transcript in wild-type A. thaliana roots, leaves and inflorescences is approximately 4.0kb in length, and that a similar transcript size occurs in mutant tissue 20 (data not shown).

The derived amino acid sequence of the RSW1 polypeptide encoded by the cosmid clone 23H12 (i.e. the polypeptide set forth in SEQ ID NO:6) is 1081 amino acids in length and contains the entire D, D, D, QXXRW signature characteristic of β-glycosyl transferase proteins, between amino acid position 395 and amino acid position 822. The conserved Aspartate, Glutamine, Arginine and Tryptophan residues are shown below, in bold type, with the local amino acid residues also indicated:

1. amino acid residues 391 to 399 of SEQ ID NO:6:

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2. Amino acid residues 557 to 565 of SEQ ID NO:6:

#### LLNVDCDHY;

3. Amino acid residues 776 to 784 of SEQ ID NO:6:

SVTEDILTG; and

4. Amino acid residues 814 to 826 of SEQ ID NO:6:

#### DRLNOVLRWALGS.

The second and third conserved Aspartate residues listed *supra*, and the fourth conserved amino acid sequence motif listed *supra* (i.e. QVLRW) are also present in the cDNA clone 10 T20782 (see Example 4 above).

The 23H12 clone potentially encodes a cellulose synthase.

#### 15 EXAMPLE 6

#### COMPLEMENTATION OF THE rsw1 MUTATION

The complementation of the cellulose mutant plant rswl is the key test to demonstrate the function of the clone 23H12 gene product. Complementation of the rswl phenotype was demonstrated by transforming the binary cosmid clone 23H12, or a derivative clone thereof encoding a functional gene product, into the Arabidopsis thaliana cellulose mutant rswl. Two DNA constructs (23H12 and pRSW1) were used to complement the rswl mutant plant line.

#### 25 1. Construct 23H12

5

Clone 23H12 is described in Example 5 and Figure 5.

#### 2. Construct pRSW1

The 23H12 construct has an insert of about 21kb in length. To demonstrate that any 30 complementation of the phenotype of the rsw1 mutation is the result of expression of the gene

which corresponds to SEQ ID NO:3, a genetic construct, designated as pRSW1, comprising the putative RSW1 gene with most of the surrounding DNA deleted, was produced. A restriction enzyme (RE) map of the RSW1 gene insert in pRSW1 is provided in Figure 5.

5 To produce pRSW1, the RSW1 gene was subcloned from cosmid 23H12 and cloned into the binary plasmid pBIN19. Briefly, Escherichia coli cells containing cosmid 23H12 were grown in LB medium supplemented with tetracyclin (3.5 mg/L). Plasmid DNA was prepared by alkaline lysis and digested sequentially with restriction enzymes PvuII and SalI. Two co-migrating fragments of 9 kb and 10 kb, respectively, were isolated as a single fraction from a 0.8% (w/v) agarose gel. The RSW1 gene was contained on the 10 kb PvuII/SalI fragment. The 9 kb fragment appeared to be a PvuII cleavage product not comprising the RSW1 gene. The restriction fragments were ligated into pBIN19 digested with Smal and SalI. An aliquot of the ligation mix was introduced by electroporation into E.coli strain XLB1. Colonies resistant to kanamycin (50 mg/L) were selected and subsequently characterised by restriction enzyme analysis to identify those clones which contained only the 10 kb PvuII/SalI fragment comprising the RSW1 gene, in pBIN19.

#### 3. Transfer of the 23HI2 and pRSW1 constructs to Agrobacterium tumefaciens

Cosmid 23H12 was transferred to Agrobacterium by triparental mating, essentially as described by Ditta et al. (1980). Three bacterial strains as follows were mixed on solid LB medium without antibiotics: Strain 1 was an E. coli helper strain containing the mobilising plasmid pRK2013, grown to stationary phase; Strain 2 was E.coli containing cosmid 23H12, grown to stationary phase; and Strain 3 was an exponential-phase culture of A. tumefaciens strain AGL1 (Lazo et al., 1991). The mixture was allowed to grow over night at 28°C, before an aliquot was streaked out on solid LB medium containing antibiotics (ampicillin 50 mg/L, rifampicin 50 mg/L, tetracyclin 3.5 mg/L) to select for transformed A. tumefaciens AGL1. Resistant colonies appeared after 2-3 days at 28°C and were streaked out once again on selective medium for further purification. Selected colonies were then subcultured in liquid LB medium supplemented with rifampicin (50 mg/L) and tetracyclin (3.5 mg/L) and stored at -80°C.

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Plasmid pRSW1 (initially designated as p2029) was introduced into A. tumefaciens strain AGL1 by electroporation.

#### 4. Transformation of rswl plants

5 The rswl plant line was transformed with constructs 23H12 and pRSW1 using vacuum infiltration essentially as described by Bechtold et al. (1993).

#### 5. Analysis of radial swelling in transformants

Complementation of the radial swelling (rsw) phenotype, which is characteristic of the rsw1 mutant plant, was assayed by germinating transformed (i.e. T1 seed) rsw1 seeds obtained as described supra on Hoaglands plates containing 50µg/ml kanamycin. Plates containing the transformed seeds were incubated at 21°C for 10-12 days. Kanamycin-resistant seedlings were transferred to fresh Hoaglands plates containing 50µg/ml kanamycin and incubated at 31°C for 2 days. Following this incubation, the root tip was examined for a radial swelling phenotype. Under these conditions, the roots of wild-type plants do not show any radial swelling phenotype however, the roots of rsw1 plants show clear radial swelling at the root tip and also have a short root compared to the wild-type plants. As a consequence, determination of the radial swelling phenotype of the transformed plants was indicative of successful complementation of the rsw1 phenotype.

20

The kanamycin-resistant seedlings were maintained by further growth of seedlings at 21 °C, following the high temperature incubation. Once plants had recovered, the seedlings were transferred to soil and grown in cabinets at 21 °C (16 hr light/8 hr dark cycle). T2 seed was then harvested from mature individual plants.

25

Using the 23H12 construct for *rswl* transformation, a total of 262 kanamycin-resistant seedlings were obtained. All of these transformants were tested for complementation of the root radial swelling phenotype. A total of 230 seedlings showed a wild type root phenotype, while only 32 seedlings showed the radial swelling root phenotype characteristic of *rswl* plants. By way of example, Figure 6 shows the phenotypes of transformed seedlings compared

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to untransformed wild-type and rsw1 seedlings, following incubation at 31°C. As shown in Figure 6, there is clear complementation of the radial swelling phenotype in the transformed seedlings, with normal root length being exhibited by the transformed seedlings at 31°C

5 Using the pRSW1 construct for transformation, a total of 140 kanamycin-resistant seedlings were obtained. All of the 11 seedlings tested for complementation of the root radial swelling phenotype showed a wild type root phenotype and none of the seedlings showed any signs of radial swelling in the roots (data not shown).

#### 10 6. General morphological analysis of the complemented rswl mutant line

Further characterisation of the complemented rswl plants has shown that other morphological characteristics of rswl have also been restored in the transgenic lines, for example the bolt (inflorescence) height, and the ability of the plants to grow wild type cotyledons, leaves, trichomes, siliques and flowers at 31 °C (data not shown).

15

#### 7. Biochemical complementation of the rswl mutant line

T2 seed from transformations using cosmid 23H12 as described supra or alternatively, using the binary plasmid pBin19 which lacks any RSW1 gene sequences, was sown on Hoagland's solid media containing kanamycin (50µg/ml), incubated for 2 days at 21°C and then transferred to 31°C for 5 days. Wild-type A.thaliana Columbia plants were grown under similar conditions but without kanamycin in the growth medium. Kanamycin resistant T2 seedlings which have at least one copy of the 23H12 cosmid sequence, and wild-type seedlings, were collected and frozen for cellulose analysis.

25 Cellulose levels were determined as acetic-nitric acid insoluble material (Updegraph, 1969) for 10 lines of kanamycin-resistant T2 plants transformed with the 23H12 cosmid sequence, and compared to the cellulose levels in rswl mutant plants, wild-type A.thaliana Columbia plants and A.thaliana Columbia plants transformed with the binary plasmid pBin19. The results are provided in Table 7.

As shown in Table 7, the cellulose levels have been significantly elevated in the complemented rswl (T2) plants, compared to the cellulose levels measured in the rswl mutant parent plant. In fact, cellulose levels in the 23H12-transformed plants, expressed relative to the fresh weight of plant material or on a per seedling basis, are not significantly different from the cellulose levels of either wild-type Arabidopsis thaliana Columbia plants or A.thaliana Columbia transformed with the binary plasmid pBin19. These data indicate that the 23H12 cosmid is able to fully complement the cellulose-deficient phenotype of the rswl mutant.

Homozygous T3 lines are generated to confirm the data presented in Table 7.

10

Furthermore, data presented in Table 7 indicate that there is no difference in the rate of growth of the T2 transformed rsw1 plants and wild-type plants at 31°C, because the fresh weight of such plants does not differ significantly. In contrast, the fresh weight of mutant rsw1 seedlings grown under identical conditions is only approximately 55% of the level observed in T2 lines transformed with 23H12 (range about 30% to about 80%). These data support the conclusion that cellulose levels have been manipulated in the complemented rsw1 (T2) plants.

Furthermore, the rate of cellulose synthesis in 23H12-transformed plants and wild-type 20 plants at 31°C, as measured by <sup>14</sup>C incorporation is also determined.

Furthermore, the  $\beta$ -1,4-glucan levels and starch levels in the 23H12 transformant lines are shown to be similar to the  $\beta$ -1,4-glucan and starch levels in wild-type plants.

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TABLE 7
CELLULOSE LEVELS IN rsw1 PLANTS TRANSFORMED
WITH COSMID CLONE 23H12

5

	PLANT LINE	SAMPLE SIZE (No. of plants)	SEEDLING FRESH WEIGHT (mg)	CELLULOSE (mg cellulose/ 100 mg tissue)	CELLULOSE (mg cellulose/ seedling)
	1.2 (rsw1+23H12)	126	2.51	1.23	0.031
10	1.4 (rsw1+23H12)	132	2.25	2.50	0.056
	2.1 (rsw1+23H12)	126	3.23	1.29	0.042
	3.1 (rsw1+23H12)	127	3.75	1.23	0.046
-	3.10 (rsw1+23H12)	128	3.52	1.69	0.060
15	4.4 (rsw1+23H12)	110	5.14	1.31	0.067
	4.5 (rsw1+23H12)	125	3.18	1.26	0.040
	5.3 (rsw1+23H12)	124	2.77	1.17	0.032
	9.2 (rsw1+23H12)	125	2.26	1.41	0.032
20	10.8 (rsw1+23H12)	126	2.4	1.20	0.029
	Columbia/pBin19	106	2.64	1.34	0.035
	Columbia	178	2.73	1.18	0.032
	rswl mutant	179	1.77	0.84	0.015

#### **EXAMPLE 7**

# DETERMINATION OF THE FULL-LENGTH NUCLEOTIDE SEQUENCE ENCODING THE WILD-TYPE RSW1 POLYPEPTIDE

5 Arabidopsis thaliana double-stranded cDNA and cDNA libraries were prepared using the CAPFINDER cDNA kit (Clontech). RNA was isolated from wild-type Columbia grown in sterile conditions for 21 days.

Approximately 100,000 cDNA clones in an unamplified cDNA library were screened under standard hybridization conditions at 65°C, using a probe comprising <sup>32</sup>P-labelled DNA amplified from double stranded cDNA. To prepare the hybridization probe, the following amplification primers were used:

- 1. 2280-F:5'GAATCGGCTACGAATTTCCCA 3'
- 2. 2370-F:5'TTGGTTGCTGGATCCTACCGG 3'
- 15 3. csp1-R:5'GGT TCT AAA TCT TCT TCC GTC 3'

wherein the primer combinations were either 2280-F/csp1-R or 2370-F/csp1-R. The primer 2280-F corresponds to nucleotide positions 2226 to 2246 in SEQ ID NO:3, upstream of the translation start site. The primer 2370-F corresponds to nucleotide positions 2314 to 2334 in SEQ ID NO:3, encoding amino acids 7 through 13 of the RSW1 polypeptide. The primer csp1-R comprises nucleotide sequences complementary to nucleotides 588 to 608 of the T20782 clone (SEQ ID NO:1) corresponding to nucleotides 6120 to 6140 of SEQ ID NO:3. The hybridization probes produced are approximately 1858 nucleotides in length (2280-F/csp1-R primer combination) or 1946 nucleotides in length (2370-F/csp1-R primer 25 combination).

Five hybridizing bacteriophage clones were identified, which were plaque-purified to homogeneity during two successive rounds of screening. Plasmids were rescued from the positively-hybridizing bacteriophage clones, using the Stratagene excision protocol for the ZapExpress<sup>TM</sup> vector according to the manufacturer's instructions. Colony hybridizations

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confirmed the identity of the clones.

Isolated cDNA clones were sequenced by primer walking similar to the method described in Examples 4 and 5 supra.

5

A full-length wild-type RSW1 nucleotide sequence was compiled from the nucleotide sequences of two cDNA clones. First, the 3'-end of the cDNA, encoding amino acids 453-1081 of RSW1, corresponded to the nucleotide sequence of the EST clone T20782 (SEQ ID NO:1). The remaining cDNA sequence, encoding amino acids 1-654 of RSW1, was 10 generated by amplification of the 5'-end from cDNA, using primer 2280-F, which comprises nucleotide sequences approximately 50-70bp upstream of the RSW1 translation start site in cosmid 23 H12, and primer csp1-R, which comprises nucleotide sequences complementary to nucleotides 588 to 608 of the T20782 clone (SEQ ID NO:1).

- 15 Several amplified clones are sequenced to show that no nucleotide errors were introduced by the amplification process. The 5' and 3' nucleotide sequences are spliced together to produce the complete RSW1 open reading frame and 3'-untranslated region provided in SEQ ID NO:5.
- 20 Those skilled in the art will be aware that the 5'-end and 3'-end of the two incomplete cDNAs are spliced together to obtain a full-length cDNA clone, the nucleotide sequence of which is set forth in SEQ ID NO:5.

Of the remaining cDNA clones, no isolated cDNA clone comprised a nucleotide sequence which precisely matched the nucleotide sequence of the RSW1 gene present in cosmid 23H12. However, several clones containing closely-related sequences were obtained, as summarised in Table 8. The nucleotide sequences of the Ath-A and Ath-B cDNAs are provided herein as SEQ ID Nos: 7 and 9, respectively.

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TABLE 8
CHARACTERISATION OF A. thaliana cDNA CLONES

	CLONE NAME	DESCRIPTION	LENGTH	SEQ ID NO:	
Ī	RSW1.1A	chimeric clone	partial	not provided	
5	RSW1A	chimeric clone	partial	not provided	
	Ath-A	12C4 cDNA	full-length	SEQ ID NO:7	
ľ	Ath-B	new sequence	full-length	SEQ ID NO:9	
	RSW4A	identical to Ath-B	full-length	not provided	

10 The derived amino acid sequences encoded by the cDNAs listed in Table 8, is provided in Figures 8 and 9 and SEQ ID Nos: 8 and 10 herein.

Figure 10 a schematic representation of the important features of the RSW1 polypeptide which are conserved within A.thaliana and between A.thaliana and other plant species. In addition to the species indicated in Figure 10, the present inventors have also identified maize, wheat, barley and Brassica ssp. cellulose biosynthetic genes by homology search. Accordingly, the present invention extends to cellulose genes and cellulose biosynthetic polypeptides as hereinbefore defined, derived from any plant species, including A. thaliana, cotton, rice, wheat, barley, maize, Eucalyptus ssp., Brassica ssp. Pinus ssp., Populus ssp., 20 Picea ssp., hemp, jute and flax, amongst others.

# EXAMPLE 8 ISOLATION OF FULL-LENGTH NUCLEOTIDE SEQUENCE ENCODING THE MUTANT RSW1 POLYPEPTIDE

25

Arabidopsis thaliana double-stranded cDNA and cDNA libraries were prepared using the CAPFINDER cDNA kit (Clontech). RNA was isolated from Arabidopsis thaliana Columbia rsw1 mutant plants grown in sterile conditions for 21 days.

30 The full-length rsw1 mutant nucleotide sequence was generated by sequencing two amplified

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DNA fragments spanning the *rsw1* mutant gene. The 5'- end sequence of the cDNA (comprising the 5'-untranslated region and exons 1-11) was amplified using the primer combination 2280-F/csp1-R (Example 7). The 3'-end sequence was amplified using the primers EST1-F and cs3-R set forth below:

1.Primer EST1-F:

5

25

5'AATGCTTCTTGTTGCCAAAGCA 3'

2.Primer cs3-R:

5'GACATGGAATCACCTTAACTGCC 3'

wherein primer EST1-F corresponds to nucleotide positions 1399-1420 of SEQ ID NO:5 (within exon 8) and primer cs3-R is complementary to nucleotides 3335-3359 of SEQ ID NO:5 (within the 3'-untranslated region of the wild-type transcript).

The full-length sequence of the mutant rsw1 transcript is set forth herein as SEQ ID NO:11.

Whilst not being bound by any theory or mode of action, a single nucleotide substitution in the rsw1 mutant nucleotide sequence (nucleotide position 1716 in SEQ ID NO:11), relative to the wild-type RSW1 nucleotide sequence (nucleotide position 1646 in SEQ ID NO:5), resulting in Ala549 being substituted with Val549 in the mutant polypeptide, may contribute to the altered activity of the RSW1 polypeptide at non-permissive temperatures such as 31°C. Additional amino acid substitutions are also contemplated by the present invention, to alter the activity of the RSW1 polypeptide, or to make the polypeptide temperature-sensitive.

# EXAMPLE 9

# ANTISENSE INHIBITION OF CELLULOSE PRODUCTION IN TRANSGENIC PLANTS

## 1. Construction of an antisense RSW1 binary vector

One example of transgenic plants in which cellulose production is inhibited is provided by the expression of an antisense genetic construct therein. Antisense technology is used to 30 target expression of a cellulose gene(s) to reduce the amount of cellulose produced by

transgenic plants.

By way of exemplification, an antisense plant transformation construct has been engineered to contain the T20782 cDNA insert (or a part thereof) in the antisense orientation and in operable connection with the CaMV 35S promoter present in the binary plasmid pRD410 (Datla et al. 1992). More particularly, the T20782 cDNA clone, which comprises the 3'-end of the wild-type RSW1 gene, was digested with XbaI and KpnI and cloned into the kanamycin-resistant derivative of pGEM3zf(-), designated as plasmid, pJKKMf(-). The RSW1 sequence was sub-cloned, in the antisense orientation, into the binary vector pRD410 as a XbaI/SacI fragment, thereby replacing the β-glucuronidase (GUS or uidA) gene. This allows the RSW1 sequence to be transcribed in the antisense orientation under the control of the CaMV 35S promoter.

The antisense RSW1 binary plasmid vector was transferred to Agrobacterium tumefaciens strain AGL1, by triparental mating and selection on rifampicin and kanamycin, as described by Lazo et al. (1991). The presence of the RSW1 insert in transformed A.tumefaciens cells was confirmed by Southern hybridization analysis (Southern, 1975). The construct was shown to be free of deletion or rearrangements prior to transformation of plant tissues, by back-transformation into Escherichia coli strain JM101 and restriction digestion analysis.

20

#### 2. Transformation of Arabidopsis thaliana

Eight pots, each containing approximately 16 A. thaliana ecotype Columbia plants, were grown under standard conditions. Plant tissue was transformed with the antisense RSW1 binary plasmid by vacuum infiltration as described by Bechtold et al (1993). Infiltration media contained 2.5% (w/v) sucrose and plants were infiltrated for 2 min until a vacuum of approximately 400mm Hg was obtained. The vacuum connection was shut off and plants allowed to sit under vacuum for 5 min.

Approximately 34,000 T1 seed was screened on MS plates containing 50µg/ml kanamycin, 30 to select for plants containing the antisense RSW1 construct. Of the T1 seed sown, 135

kanamycin-resistant seedlings were identified, of which 91 were transferred into soil and grown at 21°C under a long-day photoperiod (16hr light; 8hr dark).

Of the 91 transgenic lines, 19 lines were chosen for further analysis which had anther 5 filaments in each flower which were too short to deposit pollen upon the stigma and, as a consequence, required hand-pollination to obtain T2 seed therefrom.

T2 seed from 14 of these 19 lines was plated out onto vertical Hoaglands plates containing kanamycin to determine segregation ratios. Between five and ten seed were plated per transgenic line. Control seeds, including A. thaliana Columbia containing the binary vector pBIN19 (Bevan, 1984) and segregating 3:1 for kanamycin resistance, and the rswl mutant transformed with the NPTII gene, also segregating 3:1 for kanamycin resistance, were grown under the same conditions. Kanamycin-resistant plants were transferred to soil and grown at 21°C under long days, until flowering. Untransformed Arabidopsis thaliana Columbia plants were also grown under similar conditions, in the absence of kanamycin.

#### 3. Morphology of antisense- RSW1 plants

A comparison of the morphology of antisense RSW1 plants grown at 21°C, to mutant rswl plants grown at the non-permissive temperature (i.e. 31°C) has identified a number of common 20 phenotypes. For example, the antisense plants exhibit reduced fertility, inflorescence shortening and have short anthers, compared to wild-type plants, when grown at 21°C. These phenotypes are also observed in mutant rswl plants grown at 31°C. These results suggest that the antisense construct in the transgenic plants may be targeting the expression of the wild-type RSW1 gene at 21°C.

25

Figure 7 shows the reduced inflorescence (bolt) height in antisense 35S-RSW1 plants compared to wild-type A. thaliana Columbia plants grown under identical conditions.

#### 4. Cell wall carbohydrate analysis of antisense plants.

30 T3 plants which are homozygous for the 35S-RSW1 antisense construct are generated and the

content of cellulose therein is determined as described in Example 1. Plants expressing the antisense construct are shown to have significantly less cellulose in their cell walls, compared to wild-type plants. Additionally, the levels of non-crystalline β-1,4-glucan and starch are elevated in the cells of antisense plants, compared to otherwise isogenic plant lines which have not been transformed with the antisense genetic construct.

### 5. Antisense 35S-RSW1 mRNA expression levels in transgenic plants

Total RNA was extracted from 0.2g of leaf tissue derived from 33 kanamycin-resistant T1 plants containing the antisense 35S-RSW1 genetic construct, essentially according to 10 Longemann et al. (1986). Total RNA (25 μg) was separated on a 2.2M formaldehyde/agarose gel, blotted onto nylon filters and hybridized to a riboprobe comprising the sense strand sequence of the cDNA clone T20782. To produce the riboprobe, T7 RNA polymerase was used to transcribe sense RNA from a linearised plasmid template containing T20782, in the presence of [α-12P]UTP. Hybridizations and subsequent washes were performed as described by Dolferus et al. (1994). Hybridized membranes were exposed to Phosphor screens (Molecular Dynamics, USA).

The levels of expression of the RSW1 antisense transcript were determined and compared to the level of fertility observed for the plant lines. As shown in Table 9, the level of antisense gene expression is correlated with the reduced fertility phenotype of the antisense plants. In 13 lines, a very high or high level of expression of the 35S-RSW1 antisense gene was observed and, in 11 of these lines fertility was reduced. Only lines 2W and 3E which expressed high to very high levels of antisense mRNA, appeared to be fully fertile. In 12 lines which expressed medium levels of antisense mRNA, approximately one-half were fertile and one-half appeared to exhibit reduced fertility. In contrast, in 8 plant lines in which only a low or very low level of expression of the antisense 35S-RSW1 genetic construct was observed, a wild-type (i.e. fertile) phenotype was observed for all but one transgenic line, line 2R.

Data presented in Table 9 and Figure 7 indicate that the phenotype of the cellulose-deficient 30 mutant rsw1 may be reproduced by expressing antisense RSW1 genetic constructs in transgenic

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plants.

To confirm reduced cellulose synthesis and/or deposition in transgenic plants expressing the antisense RSW1 gene, the level of cellulose is measured by the <sup>14</sup>C incorporation assay or as acetic/nitric acid insoluble material as described in Example 1 and compared to cellulose production in otherwise isogenic wild-type plants. Cellulose production in the transgenic plants is shown to be significantly reduced compared to wild-type plants. The severity of phenotype of the transgenic plants thus produced varies considerably, depending to some extent upon the level of inhibition of cellulose biosynthesis.

10

#### TABLE 9

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# LEVELS OF ANTISENSE GENE EXPRESSION AND FERTILITY IN T1 LINES OF ANTISENSE 35S-RSW1 PLANTS

5	TI PLANT LINE	ANTISENSE 35S- <i>RSW</i> 1 EXPRESSION	FERTILITY	T1 PLANT LINE	ANTISENSE  35S- <i>RSW</i> 1  EXPRESSION	FERTILITY
	В	very high	sterile*	2H	medium	fertile
	2B	very high	sterile*	С	medium	sterile*
	3E	very high	fertile	F	medium	sterile*
10	2E	high	sterile*	2Q	medium	fertile
	2K	high	sterile*	3P	medium	sterile*
	2M	high	sterile*	3T	medium	fertile
	20	high	sterile*	5D	medium	sterile*
	2P	high	sterile*	6A	medium	fertile
15	2W	high	fertile	8E	low	fertile
	2Z	high	sterile*	2R	low	sterile*
	3G	high	sterile*	7A	low	fertile
	3Q	high	sterile*	<b>7</b> S	low	fertile
	7Q	high	sterile*	70	low	fertile
20	7N	medium	sterile*	7R	low	fertile
	7G	medium	fertile	1B	very low	fertile
	1C	medium	sterile*	<b>2</b> U	very low	fertile
	2X	medium	sterile*			

<sup>\*</sup>sterile phenotype not indicative of complete sterility, but that hand pollination at least, is

<sup>25</sup> required to obtain seed from such plants.

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# EXAMPLE 10 RSW1 RELATED SEQUENCES IN RICE PLANTS

To identify RSW1 related nucleotide sequences in rice, a genetic sequence database was searched for nucleotide sequences which were closely-related to one or more of the Arabidopsis thaliana RSW1 nucleotide sequences described in the preceding Examples. Rice EST S0542 (MAFF DNA bank, Japan) was identified, for which only a partial nucleotide sequences was available. Additionally, before the instant invention, there was no probable function attached to the rice EST S0542 sequence.

10

The present inventors have obtained the complete nucleotide sequence of clone S0542 and derived the amino acid sequence encoded therefor. The S0542 cDNA is only 1741bp in length and appears to be a partial cDNA clone because, although it comprises 100bp of 5'-untranslated sequence and contains the ATG start codon, it is truncated at 3'-end and, as a consequence encodes only the first 547 amino acid residues of the rice RSW1 or RSW1-like polypeptide. Based upon the length of the corresponding *Arabidopsis thaliana* RSW1 polypeptide (1081 amino acids), the rice RSW1 sequence set forth in SEQ ID NO:14 appears to contain approximately one-half of the complete amino acid sequence.

The N-terminal half of the rice RSW1 amino acid sequence is approximately 70% identical to the Arabidopsis thaliana RSW1 polypeptide set forth in SEQ ID NO:6, with higher homology (approximately 90%) occurring between amino acid residues 271-547 of the rice sequence. These data strongly suggest that S0542 is the rice homologue of the A. thaliana RSW1 gene. Alignments of rice, A. thaliana and cotton RSW1 amino acid sequences are presented in 25 Figures 9 and 10.

To isolate full-length cDNA clones and genomic clone equivalents of S0542 (this study and MAFF DNA bank, Japan) or D48636 (Pear et al., 1996), cDNA and genomic clone libraries are produced using rice mRNA and genomic DNA respectively, and screened by hybridisation 30 using the S0542 or D48636 cDNAs as a probe, essentially as described herein. Positive-

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hybridising plaques are identified and plaque-purified, during further rounds of screening by hybridisation, to single plaques.

The rice clones are sequenced as described in the preceding Examples to determine the complete nucleotide sequences of the rice RSW1 genes and derived amino acid sequences therefor. Those skilled in the art will be aware that such gene sequences are useful for the production of transgenic plants, in particular transgenic cereal plants having altered cellulose content and/or quality, using standard techniques. The present invention extends to all such genetic sequences and applications therefor.

10

# EXAMPLE 11 RSW1 RELATED SEQUENCES IN COTTON PLANTS

15 A <sup>32</sup>P-labelled RSW1 PCR fragment was used to screen approximately 200,000 cDNA clones in a cotton fibre cDNA library. The RSW1 PCR probe was initially amplified from Arabidopsis thaliana wild type cDNA using the primers 2280-F and csp1-R described in the preceding Examples, and then re-amplified using the primer combination 2370-F/csp1-R, also described in the preceding Examples.

20

Hybridisations were carried out under low stringency conditions at 55°C.

Six putative positive-hybridising plaques were identified in the first screening round. Using two further rounds of screening by hybridisation, four of these plaques were purified to single plaques. Three plaques hybridise very strongly to the RSW1 probe while the fourth plaque hybridises less intensely.

We conclude that the positive-hybridising plaques which have been purified are strong candidates for comprising cotton RSW1 gene sequences or RSW1-like gene sequences.

30 Furthermore, the cotton cDNAs may encode the catalytic subunit of cellulose synthase,

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because the subunit protein architecture of cellulose synthase appears to be highly conserved among plants as highlighted in the preceding Example.

Furthermore, a Southern blot of cotton genomic DNA digested with BgIII was hybridised with 5 the 5' end of the RSW1 cDNA, under low stringency hybridisation conditions at 55°C. Results are presented in Figure 11. These data demonstrate that RSW1-related sequences exist in the cotton genome.

The cotton cDNA clones described herein are sequenced as described in the preceding 10 Examples and used to produce transgenic cotton plants having altered fibre characteristics. The cDNAs are also used to genetically alter the cellulose content and/or quality of other plants, using standard techniques.

# EXAMPLE 12 RSW1 RELATED SEQUENCES IN EUCALYPTUS SSP.

Putative Eucalyptus ssp. cellulose synthase catalytic subunit gene fragments were obtained by amplification using PCR. DNA primers were designed to conserved amino acid residues found in the Arabidopsis thaliana RSW1 and 12C4 amino acid sequences. Three primers were used 20 for PCR. The primers are listed below:

pcsF-I 5'- A A/G A A G A T I G A C/T T A C/T C/T T I A A A/G G A C/T A A-3'
pcsR-II 5'-A T I G T I G G I G T I C G/T A/G T T C/T T G A/T/G/C C T/G A/T/C/G C C -3'
pcsF-II 5'- G C I A T G A A A/G A/C G I G A I T A C/T G A A/G G A -3'

25

15

Using standard PCR conditions (50°C annealing temperature) and solutions, the primer sets pcsF-I/pcsR-II and pcsF-II/pcsR-II were used to amplify genetic sequences from pooled *Eucalyptus ssp.* cDNA. In the first reaction primers pcsF-I and pcsR-II were used to generate a fragment approximately 700 bp in length. In the second PCR reaction, which used primers pcsF-II and pcsR-II, a fragment estimated to 700 bp was obtained. The sizes of the PCR

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fragments are within the size range estimated for the corresponding Arabidopsis thaliana sequences.

We conclude that the amplified *Eucalyptus ssp.* PCR fragments are likely to be related to the 5 *Arabidopsis thaliana RSW*1 gene and may encode at least a part of the *Eucalyptus ssp.* cellulose synthase catalytic subunit.

The Eucalyptus ssp. PCR clones described herein are sequenced as described in the preceding Examples and used to isolate the corresponding full-length Eucalyptus ssp cDNAs and genomic gene equivalents. Those skilled in the art will be aware that such gene sequences are useful for the production of transgenic plants, in particular transgenic Eucalyptus ssp plants having altered cellulose content and/or quality, using standard techniques. The present invention extends to all such genetic sequences and applications therefor.

15

## EXAMPLE 13

# NON-CRYSTALLINE B-1,4-GLUCAN AS A MODIFIER OF CELL WALL PROPERTIES

20 The properties of plant cell walls depend on the carbohydrates, proteins and other polymers of which they are composed and the complex ways in which they interact. Increasing the quantities of non-crystalline β-1,4-glucan in cell walls affects those wall properties which influence mechanical, nutritional and many other qualities as well as having secondary consequences resulting from the diversion of carbon into non-crystalline glucan at the expense of other uses. To illustrate one of these effects, we investigated the ability of the non-crystalline glucan to hydrogen bond to other wall components particularly cellulose in the way that has been shown to be important for wall mechanics.

Hemicelluloses such as xyloglucans cross-link cellulose microfibrils by hydrogen bonding to 30 the microfibril surface (Levy et al., 1991). Since the  $\beta$ -1,4-glucan backbone of xyloglucan is

thought to be responsible for hydrogen bonding (with the xylose, galactose and fucose substitutions limiting the capacity to form further hydrogen bonds) we can expect the non-crystalline β-1,4-glucan also to have a capacity to hydrogen bond and cross link cellulose. The effectiveness of strong alkalis in extracting xyloglucans is thought to relate to their disruption of the hydrogen bonds with cellulose (Hayashi and MacLachlan, 1984).

To demonstrate that the non-crystalline β-1,4-glucan forms similar associations with the cellulose microfibrils, we examined whether the 4 M KOH fraction, extracted from shoots of the rsw1 mutant and from wild type RSW1 plants, contained non-crystalline glucan in addition to xyloglucan. The non-crystalline glucan was separated from xyloglucan in the 4 M KOH extract by dialysing the neutralised extract against distilled water and centrifuging at 14000 g for 1 hour. The pellet was shown to be a pure β-1,4-glucan by using the methods for monosaccharide analysis, methylation analysis and enzyme digestion used to characterise the glucan in the ammonium oxalate fraction (see Example 1).

15

Table 10 shows the presence of substantial quantities of glucan recovered in pure form in the pellet from 4 M KOH fractions extracted from the overproducing rsw1 mutant of Arabidopsis thaliana. These data also demonstrate the presence of smaller quantities of non-crystalline β-1,4-glucan in the 4 M KOH fraction from wild type plants, compared to rsw1, particularly 20 when grown at 31 °C.

TABLE 10

Glucose contents\* of 4M KOH fractions from shoots of wild-type and rsw1 mutant Arabidopsis thaliana plants

	Glucose fraction	wild	-type	rsw1 r	nutant
		21°C	31°C	21°C	31°C
25	xyloglucan and non-crystalline glucan in whole extract	36.4	56.9	27.1	93.1
	non-crystalline glucan in pellet	7.8	20.5	7.6	56.0

<sup>\*,</sup> nmol glucose/ mg plant dry weight after TFA hydrolysis

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The monosaccharide composition of the supernatant remaining after centrifugation was determined after TFA hydrolysis. These data, and data from methylation analysis, are consistent with the supernatant being a relatively pure xyloglucan. The supernatant was free of glucan, because no glucose could be released by the endocellulase/β-glucosidase mixture that released glucose from β-1,4-glucan.

The presence of both non-crystalline  $\beta$ -1,4-glucan and xyloglucan in the 4 M KOH fraction, when taken together with the implications from structural predictions (Levy *et al*, 1991), is consistent with some of the non-crystalline  $\beta$ -1,4-glucan in the wall hydrogen bonding to cellulose microfibrils in similar fashion to the  $\beta$ -1,4-glucan backbone of xyloglucan.

The cross linking provided when xyloglucans and other hemicelluloses bind to two or more microfibrils is an important determinant of the mechanical properties of cellulosic walls (Hayashi, 1989). The effects of increasing the amounts of non-crystalline β-1,4-glucan in walls are likely to be greatest in walls which otherwise possess relatively low levels of cross linking as a result of high ratios of cellulose: hemicelluloses. Such conditions are common in secondary walls including those of various fibres, and the cellulose:hemicellulose ratio is particularly high in cotton fibres.

20 The effects on wall mechanical properties of overproducing non-crystalline glucan are shown by transforming plants with the mutant allele of rswl (SEQ ID NO:11) operably under the control of either the RSW1 promoter derived from SEQ ID NO:3 or SEQ ID NO:4 or alternatively, an appropriate constitutive promoter such as the CaMV 35S promoter. Production of non-crystalline glucan is quantified by fractionating the cell walls using the methods described above to show in particular that non-crystalline glucan is recovered in the 4 M KOH fraction. Mechanical properties of the cell walls are measured using standard methods for fibre analysis to study parameters such as stress-strain curves, and breaking strain, amongst other properties.

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# EXAMPLE 14 OVER-EXPRESSION OF CELLULOSE SYNTHASE IN TRANSGENIC PLANTS

5 Three strategies are employed to over-express cellulose synthase in *Arabidopsis thaliana* plants.

In the first strategy, the CaMV 35S promoter sequence is operably connected to the full-length cellulose synthase cDNA which is obtainable by primer extension of SEQ ID NO:1. This is achievable by cloning the full-length cDNA encoding cellulose synthase, in the sense orientation, between the CaMV 35S promoter or other suitable promoter operable in plants and the nopaline synthase terminator sequences of the binary plasmid pBI121.

In the second strategy, the coding part of the genomic gene is cloned, in the sense orientation, between the CaMV 35S promoter and the nopaline synthase terminator sequences of the binary plasmid pBI121.

In the third strategy, the 23H12 binary cosmid clone or the derivative pRSW1, containing the cellulose synthase gene sequence operably under the control of the cellulose synthase gene promoter and terminator sequences is prepared in a form suitable for transformation of plant tissue.

For Agrobacterium-mediated tissue transformation, binary plasmid constructs discussed supra are transformed into Agrobacterium tumefaciens strain AGL1 or other suitable strain. The recombinant DNA constructs are then introduced into wild type Arabidopsis thaliana plants (Columbia ecotype), as described in the preceding Examples.

Alternatively, plant tissue is directly transformed using the vacuum infiltration method described by Beshtold et al. (1993).

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The transgenic plants thus produced exhibit a range of phenotypes, partly because of position effects and variable levels of expression of the cellulose synthase transgene.

Cellulose content in the transgenic plants and isogenic untransformed control plants is determined by the <sup>14</sup>C incorporation assay or as acetic/nitric acid insoluble material as described in Example 1. In general, the level of cellulose deposition and rates of cellulose biosynthesis in the transgenic plants are significantly greater than for untransformed control plants.

10 Furthermore, in some cases, co-supression leads to mimicry of the rswl mutant phenotype.

# EXAMPLE 15 SITE-DIRECTED MUTAGENESIS OF THE RSW1 GENE

15

The nucleotide sequence of the RSW1 gene contained in 23H12 is mutated using site-directed mutagenesis, at several positions to alter its catalytic activity or substrate affinity or glucan properties. In one example, the RSW1 gene is mutated to comprise one or more mutations present in the mutant rsw1 allele.

20

The mutated genetic sequences are cloned into binary plasmid described in the preceding Examples, in place of the wild-type sequences. Plant tissue obtained from both wild-type Arabidopsis thaliana (Columbia) plants and A. thaliana rswl plants is transformed as described herein and whole plants are regenerated.

25

Control transformations are performed using the wild-type cellulose synthase gene sequence.

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#### **EXAMPLE 16**

### PHENOTYPES OF PLANTS EXPRESSING MUTATED RSW1 GENES

Plants transformed with genetic constructs described in Example 15 (and elsewhere) are categorised initially on the basis of number of transgene copies, to eliminate variability arising therefrom. Plants expressing single copies of different transgenes are analysed further for cell wall components, including cellulose, non-crystalline β-1,4-glucan polymer, starch and carbohydrate content.

#### 10 1. Cellulose content

Cellulose content in the transgenic plants is determined by the <sup>14</sup>C incorporation assay as described in Example 1. Cell walls are prepared, fractionated and the monosaccharide composition of individual fractions determined as in Example 1.

### 15 2. Non-crystalline β-1,4-glucan content

Transgenic plants expressing the rsw1 mutant allele exhibit a higher level of non-crystalline, and therefore extractable,  $\beta$ -1,4-glucan in cell walls compared to plants expressing an additional copy of the wild-type RSW1 allele. Thus, it is possible to change the crystallinity of the  $\beta$ -1,4-glucan chains present in the cell wall by mutation of the wild-type RSW1 allele.

20

#### 3. Starch content

Transgenic plants are also analysed to determine the effect of mutagenesis of the RSW1 gene on the level of starch deposited in their roots. The quantity of starch present in material prepared from the crude wall fraction is determined using the anthrone/H<sub>2</sub>SO<sub>4</sub> method described in Example 1. The data show that mutating the RSW1 gene to the mutant rsw1 allele increases starch deposition. This demonstrates that the gene can be used to alter the partitioning of carbon into carbohydrates other than cellulose.

### 4.Cell wall composition

30 The cell wall composition of transgenic plant material is also analysed. Wild type and rswl

and transgenic seedlings are grown for 2 d at 21°C and then kept for a further 5 d at either 21°C or 31°C. With transfer to 31°C when the seed has scarcely germinated, the wall composition at final harvest largely reflects the operation of the mutated rsw1 gene product at its restrictive temperature. Cell wall fractionation is carried out in similar fashion to that described for the <sup>14</sup>C-experiment (Example 1) and the monosaccharide composition of each fraction is quantified by GC/MS after hydrolysis with trifluoroacetic acid or, in the case of crystalline cellulose, H<sub>2</sub>SO<sub>4</sub>.

In some transgenic plants in which the RSW1 gene is mutated, the monosaccharide 10 composition is comparable to that observed for homozygous rsw1 plants, at least in some cases, confirming that there is a major reduction in the quantity of crystalline cellulose in the final, acid insoluble fraction. Thus, mutation of the RSW1 gene can be performed to produce changes in the composition of plant cell walls.

### 15 EXAMPLE 17

# CHEMICAL MODIFICATION OF THE RSWI GENE TO MANIPULATE CELLULOSE PRODUCTION AND PLANT CELL WALL CONTENT.

As demonstrated in the preceding Examples, the RSW1 gene is involved in cellulose 20 production and the manipulation of cell wall content.

In the present Example, to identify novel phenotypes and gene sequences important for the normal functioning of the cellulose synthase gene, the RSW1 gene is modified in planta, using the chemical mutagen EMS. The mutant plants are identified following germination and the modified RSW1 genes are isolated and characterised at the nucleotide sequence level. A sequence comparison between the mutant gene sequences and the wild type sequence reveals nucleotides which encode amino acids important to the normal catalytic activity of the cellulose synthase enzyme, at least in Arabidopsis thaliana plants.

30 This approach thus generates further gene sequences of utility in the modification of cellulose

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content and properties in plants.

# **EXAMPLE 18 DISCUSSION**

5

Five pieces of evidence make a compelling case that the RSW1 gene product encodes the catalytic subunit of cellulose synthase:

- 1. The rsw1 mutation selectively inhibits cellulose synthesis and promotes accumulation of a non-crystalline  $\beta$ -1,4-glucan;
- 10 2. The rsw1 mutation removes cellulose synthase complexes from the plasma membrane, providing a plausible mechanism for reduced cellulose accumulation and placing the RSW1 product either in the complexes or interacting with them;
  - 3. The D,D,D,QXXRW signature identifies the RSW1 gene product as a processive glycosyl transferase enzyme (Saxena, 1995);
- 15 4. The wild type allele corrects the temperature sensitive phenotype of the rsw1 mutant; and
  - 5. Antisense expression of the RSW1 in transgenic plants grown at 21 °C reproduces some of the phenotype of rsw1 which is observed following growth at 31 °C.
- 20 Consistent with the plasma membrane location expected for a catalytic subunit, the putative 122 kDa RSW1 product contains 8 predicted membrane-spanning regions. Six of these regions cluster near the C-terminus (Figure 10), separated from the other two by a domain that is probably cytoplasmic and has the weak sequence similarities to prokaryotic glycosyl transferases (Wong, 1990; Saxena, 1990; Matthyse, 1995; Sofia, 1994; Kutish, 1996).

25

RSW1 therefore qualifies as a member of the large family of *Arabidopsis thaliana* genes whose members show weak similarities to bacterial cellulose synthase. RSW1 is the first member of that family to be rigorously identified as an authentic cellulose synthase. Among the diverse genes in *A. thaliana*, at least two genes show very strong sequence similarities to 30 the *RSW*1 gene and are most likely members of a highly conserved sub-family involved in

cellulose synthesis. The closely related sequences come from cosmid 12C4, a partial genomic clone cross-hybridising with EST T20782 designated *Ath*-A, and from a full length cDNA designated *Ath*-B.

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5 Ath-A resembles RSW1 (SEQ ID NO:5) at its N-terminus whereas Ath-B starts 22 amino acid residues downstream [Figure 8 and Figure 9(i), (ii) and (iii)]. Closely related sequences in other angiosperms are the rice EST S0542 [Figure 9(i), (ii) and (iii)], which resembles the polypeptides encoded by RSW1 and Ath-A and the cotton celA1 gene (Pear, 1996) at the N-terminus.

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The Arabidopsis thaliana, rice and cotton genes have regions of very high sequence similarity interspersed with variable regions (Figures 9 and 10). Most of the highest conservation among those gene products occurs in their central cytoplasmic domain where the weak similarities to the bacterial cellulose synthase occur. The N-terminal region that precedes the first 15 membrane spanning region is probably also cytoplasmic but shows many amino acid substitutions as well as sequences in RSW1 that have no counterpart in some of the other genes as already noted for celA. An exception to this is a region comprising 7 cysteine residues with highly conserved spacings (Figure 10). This is reminiscent of regions suggested to mediate protein-protein and protein-lipid interactions in diverse proteins including 20 transcriptional regulators and may account for the striking sequence similarity between this region of RSW1 and two putative soybean bZIP transcription factors (Genbank SOYSTF1A and 1B).

In conclusion, the chemical and ultrastructural changes seen in the cellulose-deficient mutant combine with gene cloning and complementation of the mutant to provide strong evidence that the RSW1 locus encodes the catalytic subunit of cellulose synthase. Accumulation of non-crystalline β-1,4-glucan in the shoot of the rsw1 mutant suggests that properties affected by the mutation are required for glucan chains to assemble into microfibrils. Whilst not being bound by any theory or mode of action, a key property may be the aggregation of catalytic subunits into plasma membrane rosettes. At the restrictive temperature, mutant synthase

complexes disassemble to monomers (or smaller oligomers) that are undetectable by freeze etching. At least in the shoot, the monomers seem to remain biosynthetically active but their β-1,4-glucan products fail to crystallise into microfibrils probably because the chains are growing from dispersed sites. Crystallisation into microfibrils, with all its consequences for wall mechanics and morphogenesis, therefore may depend upon catalytic subunits remaining aggregated as plasma membrane rosettes.

Those skilled in the art will appreciate that the invention described herein is susceptible to variations and modifications other than those specifically described. It is to be understood that the invention includes all such variations and modifications. The invention also includes all of the steps, features, compositions and compounds referred to or indicated in this specification, individually or collectively, and any and all combinations or any two or more of said steps or features.

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#### SEQUENCE LISTING

(1) GENERAL INFORMATION: 5 (i) APPLICANT: Australian National University and the Commonwealth Scientific and Industrial Research Organisation 10 (ii) TITLE OF INVENTION: Manipulation of plant cellulose (iii) NUMBER OF SEQUENCES: 14 (iv) CORRESPONDENCE ADDRESS: 15 (A) ADDRESSEE: Davies Collison Cave Patent Attorneys (B) STREET: 1, Little Collins Street (C) CITY: Melbourne (D) STATE: Victoria (E) COUNTRY: Australia 20 (F) ZIP: 3000 (v) COMPUTER READABLE FORM: (A) MEDIUM TYPE: Floppy disk (B) COMPUTER: IBM PC compatible 25 (C) OPERATING SYSTEM: PC-DOS/MS-DOS (D) SOFTWARE: PatentIn Release #1.0, Version #1.25 (vi) CURRENT APPLICATION DATA: (A) APPLICATION NUMBER: PCT INTERNATIONAL 30 (B) FILING DATE: (vii) PRIOR APPLICATION DATA: (A) APPLICATION NUMBER: AU PO0699 (B) FILING DATE: 27-JUN-1996 35 (viii) ATTORNEY/AGENT INFORMATION: (A) NAME: SLATTERY, JOHN M (ix) TELECOMMUNICATION INFORMATION: 40 (A) TELEPHONE: 61-3-9254-2777

(B) TELEFAX: 61-3-9254-2770

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(C) TELEX: AA31787

(2) INFORMATION FOR SEQ ID NO:1:

5 (i) SEQUENCE CHARACTERISTICS:

(A) LENGTH: 2248 base pairs

(B) TYPE: nucleic acid

(C) STRANDEDNESS: single

(D) TOPOLOGY: linear

10

(ii) MOLECULE TYPE: cDNA

(iii) HYPOTHETICAL: NO

15 (vi) ORIGINAL SOURCE:

(A) ORGANISM: Arabidopsis thaliana

(vii) IMMEDIATE SOURCE:

(B) CLONE: EST T20782

20

(ix) FEATURE:

(A) NAME/KEY: CDS

(B) LOCATION: 1..1887

25

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:1:

CGA GCT ATG AAG AGA GAG TAT GAA GAG TTT AAA GTG AGG ATA AAT GCT

48

Arg Ala Met Lys Arg Glu Tyr Glu Glu Phe Lys Val Arg Ile Asn Ala

30 1 5 10 15

CTT GTT GCC AAA GCA CAG AAA ATC CCT GGA GAA GGC TGG ACA ATG CAG 96
Leu Val Ala Lys Ala Gln Lys Ile Pro Gly Glu Gly Trp Thr Met Gln

20 25 30

35

GAT GGT ACT CCC TGG CCT GGT AAC AAC ACT AGA GAT CAT CCT GGA ATG

144
Asp Gly Thr Pro Trp Pro Gly Asn Asn Thr Arg Asp His Pro Gly Met

35 40 45

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	ATA	CAG	GTG	TTC	TTA	GGC	CAT	AGT	GGG	GGT	CTG	GAT	ACC	GAT	GGA	AAT	192
	Ile	Gln	Val	Phe	Leu	Gly	His	Ser	Gly	Gly	Leu	Asp	Thr	qaA	Gly	Asn	
		50					55					60					
5	GAG	CTG	CCT	AGA	CTC	ATC	TAT	GTT	TCT	CGT	GAA	AAG	CGG	CCT	GGA	TTT	240
	Glu	Leu	Pro	Arg	Leu	Ile	Tyr	Val	Ser	Arg	Glu	Lys	Arg	Pro	Gly	Phe	
	65					70					75					80	
	CAA	CAC	CAC	AAA	AAG	GCT	GGA	GCT	ATG	TAA	GCA	TCG	ATC	CGT	GTA	TCT	288
10	Gln	His	His	Lys	Lys	Ala	Gly	Ala	Met	Asn	Ala	Ser	Ile	Arg	Val	Ser	
					85					90					95		
	GCT	GTT	CTT	ACC	AAT	GGA	GCA	TAT	CTT	TTG	AAC	GTG	GAT	TGT	GAT	CAT	336
	Ala	Val	Leu	Thr	Asn	Gly	Ala	Tyr	Leu	Leu	Asn	Val	Asp	Cys	Asp	His	
15				100					105					110			
	TAC	TTT	AAT	AAC	AGT	AAG	GCT	ATT	AAA	GAA	GCT	atg	TGT	TTC	ATG	ATG	384
	Tyr	Phe	Asn	Asn	Ser	Lys	Ala	Ile	Lys	Glu	Ala	Met	Cys	Phe	Met	Met	
			115					120					125				
20																	
	GAC	CCG	GCT	ATT	GGA	AAG	AAG	TGC	TGC	TAT	GTC	CAG	TTC	CCT	CAA	CGT	432
	Asp	Pro	Ala	Ile	Gly	Lys	Lys	Сув	Cys	Tyr	Val	Gln	Phe	Pro	Gln	Arg	
		130					135					140					
25	TTT	GAC	GGT	ATT	GAT	TTG	CAC	GAT	CGA	TAT	GCC	AAC	AGG	AAT	ATA	GTC	480
	Phe	Asp	Gly	Ile	Asp	Leu	His	qaA	Arg	Tyr	Ala	Asn	Arg	Asn	Ile	Val	
	145					150					155					160	
				ATT													528
30	Phe	Phe	Asp	Ile	Asn	Met	Lys	Gly	Leu	Asp	Gly	Ile	His	Gly	Pro	Val	
					165					170					175		
	TAT	GTG	GGT	ACT	GGT	TGT	TGT	TTT	AAT	AGG	CAG	GCT	CTA	TAT	GGG	TAT	576
	Tyr	Val	Gly	Thr	Gly	Сув	Сув	Phe	Asn	Arg	Gln	Ala	Leu	Tyr	Gly	Tyr	
35				180					185					190			
	GAT	CCT	GTT	TTG	ACG	GAA	GAA	GAT	TTA	GAA	CCA	AAT	ATT	ATT	GTC	AAG	624
	Asp	Pro	Val	Leu	Thr	Glu	Glu	Asp	Leu	Glu	Pro	Asn	Ile	Ile	Val	Lys	
			195					200					205				
40																	

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	AGC	TGT	TGC	GGG	TCA	AGG	AAG	AAA	GGT	AAA	AGT	AGC	AAG	AAG	TAT	AAC	672
	Ser	Сув	Сув	Gly	Ser	Arg	Lys	Lys	Gly	Lys	Ser	Ser	Lys	Lys	Tyr	Asn	
		210					215					220					
5	TAC	GAA	AAG	AGG	AGA	GGC	ATC	AAC	AGA	AGT	GAC	TCC	AAT	GCT	CCA	CTT	720
	Tyr	Glu	Lys	Arg	Arg	Gly	Ile	Asn	Arg	Ser	Asp	Ser	Asn	Ala	Pro	Leu	
	225					230					235					240	
	TTC	AAT	ATG	GAG	GAC	ATC	GAT	GAG	GGT	TTT	GAA	GGT	TAT	GAT	GAT	GAG	768
10	Phe	Asn	Met	Glu	Asp	Ile	qeA	Glu	Gly	Phe	Glu	Gly	Tyr	Asp	Asp	Glu	
					245					250					255		
	AGG	TCT	ATT	CTA	ATG	TCC	CAG	AGG	AGT	GTA	GAG	AAG	CGT	TTT	GGT	CAG	816
	Arg	Ser	Ile	Leu	Met	Ser	Gln	Arg	Ser	Val	Glu	Lys	Arg	Phe	Gly	Gln	
15				260					265					270			
	TCG	CCG	GTA	TTT	ATT	GCG	GCA	ACC	TTC	ATG	GAA	CAA	GGC	GGC	ATT	CCA	864
	Ser	Pro	Val	Phe	Ile	Ala	Ala	Thr	Phe	Met	Glu	Gln	Gly	Gly	Ile	Pro	
			275					280					285				
20																	
	CCA	ACA	ACC	AAT	CCC	GCT	ACT	CTT	CTG	AAG	GAG	GCT	ATT	CAT	GTT	ATA	912
	Pro	Thr	Thr	Asn	Pro	Ala	Thr	Leu	Leu	Lys	Glu	Ala	Ile	His	Val	Ile	
		290					295			•		300					
25	AGC	TGT	GGT	TAC	GAA	GAC	AAG	ACT	GAA	TGG	GGC	AAA	GAG	ATT	GGT	TGG	960
				Tyr													
	305					310					315					320	
	ATC	TAT	GGT	TCC	GTG	ACG	GAA	GAT	ATT	CTT	ACT	GGG	TTC	AAG	ATG	CAT	1008
30	Ile	Tyr	Gly	Ser	Val	Thr	Glu	Asp	Ile	Leu	Thr	Gly	Phe	Lys	Met	His	
					325					330		-		•	335		
	GCC	CGG	GGT	TGG	ATA	TCG	ATC	TAC	TGC	AAT	CCT	CCA	CGC	CCT	GCG	TTC	1056
	Ala	Arg	Gly	Trp	Ile	Ser	Ile	Tyr	Сув	Asn	Pro	Pro	Arg	Pro	Ala	Phe	
35		_	_	340				·	345					350			
	AAG	GGA	TCT	GCA	CCA	ATC	AAT	CTT	TCT	GAT	CGT	TTG	AAC	CAA	GTT	CTT	1104
	Lys	Gly	Ser	Ala	Pro	Ile	Asn	Leu	Ser	Asp	Arg	Leu	Asn	Gln	Val	Leu	
	-	-	355					360		•			365				
40																	

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	CGA	TGG	GCT	TTG	GGA	TCT	ATC	GAG	TTA	CTT	CTT	AGC	AGA	CAT	TGT	CCT	1152
	Arg	Trp	Ala	Leu	Gly	Ser	lle	Glu	Ile	Leu	Leu	Ser	Arg	His	Суз	Pro	
		370					375					380					
5	ATC	TGG	TAT	GGT	TAC	CAT	GGA	AGG	TTG	AGA	CTT	TTG	GAG	AGG	ATC	GCT	1200
	Ile	Trp	Tyr	Gly	Tyr	His	Gly	Arg	Leu	Arg	Leu	Leu	Glu	Arg	Ile	Ala	
	385					390					395					400	
	TAT	ATC	AAC	ACC	ATC	GTC	TAT	CCT	ATT	ACA	TCC	ATC	CCT	CTT	ATT	GCG	1248
10	Tyr	Ile	Asn	Thr	Ile	Val	Tyr	Pro	Ile	Thr	Ser	Ile	Pro	Leu	Ile	Ala	
					405					410					415		
	TAT	TGT	ATT	CTT	CCC	GCT	TTT	TGT	CTC	ATC	ACC	GAC	AGA	TTC	ATC	ATA	1296
	Tyr	Cys	Ile	Leu	Pro	Ala	Phe	Сув	Leu	Ile	Thr	Asp	Arg	Phe	Ile	Ile	
15				420					425					430			
	ccc	GAG	ATA	AGC	AAC	TAC	GCG	AGT	ATT	TGG	TTC	ATT	CTA	CTC	TTC	ATC	1344
	Pro	Glu	Ile	Ser	Asn	Tyr	Ala	Ser	Ile	Trp	Phe	Ile	Leu	Leu	Phe	Ile	
			435					440					445				
20																	
	TCA	ATT	GCT	GTG	ACT	GGA	ATC	CTG	AAA	CTG	AAA	TGG	AAC	GGT	GTG	AGC	1392
	Ser	Ile	Ala	Val	Thr	Gly	Ile	Leu	Lys	Leu	Lys	Trp	Asn	Gly	Val	Ser	
		450					455					460					
25	ATT	GAG	GAT	TGG	TGG	AGG	AAC	AAC	CAG	TTC	TGG	GTC	ATT	GGT	GGC	ACA	1440
	Ile	Glu	Asp	Trp	Trp	Arg	Asn	Asn	Gln	Phe	Trp	Val	Ile	Gly	Gly	Thr	
	465					470					475					480	
	TCC	ACC	CAT	CTT	TTT	GCT	GTC	TTC	CAA	GGT	CTA	CTT	AAG	GTT	CTT	GCT	1488
30	Ser	Thr	His	Leu	Phe	Ala	Val	Phe	Gln	Gly	Leu	Leu	Lys	Val	Leu	Ala	
					485					490					495		
	GGT	ATC	AAC	ACC	AAC	TTC	ACC	GTT	ACA	TCT	AAA	GCC	ACA	AAC	AAA	AAT	1536
	Gly	Ile	Asn	Thr	Asn	Phe	Thr	Val	Thr	Ser	Lys	Ala	Thr	Asn	Lys	Asn	
35				500					505					510			
	GGG	GAT	TTT	GCA	AAA	стс	TAC	ATC	TTC	AAA	TGG	ACA	GCT	CTT	CTC	ATT	1584
	Gly	Asp	Phe	Ala	Lys	Leu	Tyr	Ile	Phe	Lys	Trp	Thr	Ala	Leu	Leu	Ile	
			515					520					525				
40																	

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	CCA	CCA	ACC	ACC	GTC	CTA	CTT	GTG	AAC	CTC	ATA	GGC	ATT	GTG	GCT	GGT	1632
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		530					535					540					
5	GTC	TCT	TAT	GCT	GTA	AAC	AGT	GGC	TAC	CAG	TCG	TGG	GGT	CCG	CTT	TTC	1680
	Val	Ser	Tyr	Ala	Val	Asn	Ser	Gly	Tyr	Gln	Ser	Trp	Gly	Pro	Leu	Phe	
	545					550					555					560	
	GGG	AAG	CTC	TTC	TTC	GCC	TTA	TGG	GTT	ATT	GCC	CAT	CTC	TAC	CCT	TTC	1728
10	Gly	Lys	Leu	Phe	Phe	Ala	Leu	Trp	Val	Ile	Ala	His	Leu	Tyr	Pro	Phe	
					565					570					575		
	TTG	AAA	GGT	CTG	TTG	GGA	AGA	CAA	AAC	ÇGA	ACA	CCA	ACC	ATC	GTC	TTA	1776
	Leu	Lys	Gly	Leu	Leu	Gly	Arg	Gln	Asn	Arg	Thr	Pro	Thr	Ile	Val	Ile	
15				580					585					590			
	GTC	TGG	TCT	GTT	CTT	CTC	GCC	TCC	ATC	TTC	TCG	TTG	CTT	TGG	GTC	AGG	1824
	Val	Trp	Ser	Val	Leu	Leu	Ala	Ser	Ile	Phe	Ser	Leu	Leu	Trp	Val	Arg	
			595					600					605				
20																	
	ATC	AAT	ccc	TTT	GTG	GAC	GCC	AAT	ccc	AAT	GCC	AAC	AAC	TTC	AAT	GGC	1872
	Ile	Asn	Pro	Phe	Val	Asp	Ala	Asn	Pro	Asn	Ala	Asn	Asn	Phe	Asn	Gly	
		610					615					620					
25	AAA	GGA	GGT	GTC	TTT	TAG	ACCC	TAT :	TAT	ATAC:	rt G	rgtgt	rgca'	r at	ATCA	AAAA	1927
	Lys	Gly	Gly	Val	Phe												
	625																
	CGC	GCAA'	TGG (	GAAT'	TCCA	AA TO	CATC	AAA1	cci	ATCA:	AACC	CCAC	STGA	ACC (	GGC	AGTTAA	1987
30																	
	GGT	SATT	CCA '	TGTC	CAAG	AT TA	AGCT	rtct	C CG/	GTA	3CCA	GAG	AAGG'	rga i	AATT	GTTCGT	2047
	AAC	ACTA'	TTG '	TAAT	GATT	TT C	CAGT	3GGG	A AG	AAGA:	rgtg	GAC	CAA	ATG 2	ATAC	ATAGTC	2107
35	TAC	AAAA	AGA A	ATTA	GTTA'	TA A	TTT	CTTA:	r at:	TAT	TTA	TTT	AAAG	CTT (	GTTA(	GACTCA	2167
	CAC	TAT	GTA I	ATGT'	TGGA	AC T	rgtt	STCC:	LAA 1	AAAG	GAT	TGG	GTT	TTC :	TTTT	<b>FATCTA</b>	2227
	AGA	ATCT(	GAA (	GTTT	ATAT	GC T											2248

40

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(2)	INFORMATION	FOR	SEQ	ID	NO:2:

(i) SEQUENCE CHARACTERISTICS:

(A) LENGTH: 629 amino acids

5 (B) TYPE: amino acid

(D) TOPOLOGY: linear

(ii) MOLECULE TYPE: protein

10 (xi) SEQUENCE DESCRIPTION: SEQ ID NO:2:

Arg Ala Met Lys Arg Glu Tyr Glu Glu Phe Lys Val Arg Ile Asn Ala 1 5 10 15

15 Leu Val Ala Lys Ala Gln Lys Ile Pro Gly Glu Gly Trp Thr Met Gln
20 25 30

Asp Gly Thr Pro Trp Pro Gly Asn Asn Thr Arg Asp His Pro Gly Met
35 40 45

20

Ile Gln Val Phe Leu Gly His Ser Gly Gly Leu Asp Thr Asp Gly Asn
50 55 60

Glu Leu Pro Arg Leu Ile Tyr Val Ser Arg Glu Lys Arg Pro Gly Phe
25 65 70 75 80

Gln His His Lys Lys Ala Gly Ala Met Asn Ala Ser Ile Arg Val Ser 85 90 95

30 Ala Val Leu Thr Asn Gly Ala Tyr Leu Leu Asn Val Asp Cys Asp His

Tyr Phe Asn Asn Ser Lys Ala Ile Lys Glu Ala Met Cys Phe Met Met
115 120 125

35

Asp Pro Ala Ile Gly Lys Lys Cys Cys Tyr Val Gln Phe Pro Gln Arg 130 135 140

Phe Asp Gly Ile Asp Leu His Asp Arg Tyr Ala Asn Arg Asn Ile Val 40 145 150 155 160

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	Phe	Phe	Asp	Ile	Asn 165	Met	Lys	Gly	Leu	Asp 170	Gly	Ile	His	Gly	Pro 175	Val
5	Туr	Val	Gly	Thr 180	Gly	Сув	Cys	Phe	Asn 185	Arg	Gln	Ala	Leu	Tyr 190	Gly	Tyr
	Asp	Pro	Val 195	Leu	Thr	Glu	Glu	Asp 200	Leu	Glu	Pro	Asn	11e 205	Ile	Val	Lys
10	Ser	Cys 210	Cys	Gly	Ser	Arg	Lys 215	Lys	Gly	Lys	Ser	Ser 220	Lys	Lys	Tyr	Asn
16	Tyr 225	Glu	Lys	Arg	Arg	Gly 230	Ile	Asn	Arg	Ser	Авр 235	Ser	Asn	Ala	Pro	Leu 240
15	Phe	Авп	Met	Glu	Авр 245	Ile	Авр	Glu	Gly	Phe 250	Glu	Gly	Tyr	Asp	Авр 255	Glu
20	Arg	Ser	Ile	Leu 260	Met	Ser	Gln	Arg	Ser 265	Val	Glu	Lys	Arg	Phe 270	Gly	Gln
	Ser	Pro	Val 275	Phe	Ile	Ala	Ala	Thr 280	Phe	Met	Glu	Gln	Gly 285	Gly	Ile	Pro
25	Pro	Thr 290	Thr	Asn	Pro	Ala	Thr 295	Leu	Leu	Lys	Glu	Ala 300	Ile	His	Val	Ile
30	Ser 305	Cys	Gly	Tyr	Glu	Asp 310	Lys	Thr	Glu	Trp	Gly 315	Lys	Glu	Ile	Gly	Trp
30	Ile	Tyr	Gly	Ser	Val 325	Thr	Glu	Asp	Ile	Leu 330	Thr	Gly	Phe	Lys	Met 335	His
35	Ala	Arg	Gly	Trp 340	Ile	Ser	Ile	Tyr	Сув 345	Asn	Pro	Pro	Arg	Pro 350	Ala	Phe
	Lys	Gly	Ser	Ala	Pro	Ile	Asn	Leu 360	Ser	Asp	Arg	Leu	Asn 365	Gln	Val	Leu

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	Arg	Trp	Ala	Leu	Gly	Ser	Ile	Glu	Ile	Leu	Leu	Ser	Arg	His	Cys	Pro
		370					375					380				
		Trp	Tyr	Gly	Tyr	His	Gly	Arg	Leu	Arg	Leu	Leu	Glu	Arg	Ile	Ala
5	385					390					395					400
	Tyr	Ile	Asn	Thr	Ile	Val	Tyr	Pro	Ile		Ser	Ile	Pro	Leu	Ile	Ala
10					405					410					415	
10	Tyr	Сув	Ile		Pro	Ala	Phe	Cys		Ile	Thr	qaA	Arg		Ile	Ile
	D	<b>6</b> 3	• • • • • • • • • • • • • • • • • • • •	420	•				425	_	_,			430		
	Pro	GIU		ser	Asn	Tyr	Ala		He	Trp	Pne	He		Leu	Phe	Ile
15			435		_,			440		_	_	_	445		٨	
	Ser	11e 450	Ala	Val	Thr	Gly		Leu	Lys	Leu	Lys	_	Asn	Gly	Val	Ser
				_	_	_	455			_,	_	460				
20	11e	Glu	Asp	Trp	Trp		Asn	Asn	Gln	Phe	_	Val	Ile	Gly	Gly	
20		<b>57</b> 1	***	•		470		-1		<b>-</b> 3	475	_	_			480
	ser	Thr	HIS	Leu	Phe	Ala	Val	Phe	GIn	-	Leu	Leu	Lys	Val		Ala
25	Clu	Tlo	A a m	Th .ee	485 Asn	Dha	Th.	1r_1	mb	490	•	.1	ent		495	
23	GIY	116	Asn	500	Asn	Pne	ınr	vai	505	ser	гÀв	Ala	inr		Lys	ASI
	<b>63</b>	<b>3</b>	<b>D</b> b -		•	•		-, -		•		_1		510		
	GIY	Asp	515	Ala	ГÀЗ	Leu	Tyr	520	Pne	ràs	Trp	Thr		Leu	Leu	He
30			J. J					520					525			
	Pro	Pro	Thr	Thr	Val	Leu	Leu	Val	Asn	Leu	Tle	Glv	Tle	Val	Ala	Gla
		530					535					540				,
	Val	Ser	Tyr	Ala	Val	Asn	Ser	Gly	Tyr	Gln	Ser	Trp	Gly	Pro	Leu	Phe
35	545					550					555					560
	Gly	Lys	Leu	Phe	Phe	Ala	Leu	Trp	Val	Ile	Ala	His	Leu	Tyr	Pro	Phe
					565					570					575	

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Leu Lys Gly Leu Leu Gly Arg Gln Asn Arg Thr Pro Thr Ile Val Ile 580 585 590

Val Trp Ser Val Leu Leu Ala Ser Ile Phe Ser Leu Leu Trp Val Arg 5 595 600 605

Ile Asn Pro Phe Val Asp Ala Asn Pro Asn Ala Asn Asn Phe Asn Gly
610 615 620

 $10~{
m Lys}~{
m Gly}~{
m Gly}~{
m Val}~{
m Phe}$ 

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(2) INFORMATION FOR SEQ ID NO:3:
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(i) SEQUENCE CHARACTERISTICS:

5 (A) LENGTH: 8411 base pairs

(B) TYPE: nucleic acid(C) STRANDEDNESS: single

(D) TOPOLOGY: linear

10 (ii) MOLECULE TYPE: DNA (genomic)

(iii) HYPOTHETICAL: NO

(iv) ANTI-SENSE: NO

15

(vi) ORIGINAL SOURCE:

(A) ORGANISM: Arabidopsis thaliana

(B) STRAIN: Columbia (wild-type)

20 (vii) IMMEDIATE SOURCE:

(B) CLONE: 23H12 RSW1 GENE

(ix) FEATURE:

25 (A) NAME/KEY: exon

(B) LOCATION: 2296..2376

(ix) FEATURE:

(A) NAME/KEY: exon

30 (B) LOCATION: 2904..3099

(ix) FEATURE:

(A) NAME/KEY: exon

(B) LOCATION: 3198..3370

35

(ix) FEATURE:

(A) NAME/KEY: exon

(B) LOCATION: 3594..3708

40 (ix) FEATURE:

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(A) NAME/KEY: exon
(B) LOCATION: 3824..4013

(ix) FEATURE:

5 (A) NAME/KEY: exon
(B) LOCATION: 4181..4447

(1x) FEATURE:

(A) NAME/KEY: exon

10 (B) LOCATION: 4783..5128

(1x) FEATURE:

(A) NAME/KEY: exon

(B) LOCATION: 5207..5344

15

(ix) FEATURE:

(A) NAME/KEY: exon

(B) LOCATION: 5426..5551

20 (ix) FEATURE:

(A) NAME/KEY: exon

(B) LOCATION: 5703..5915

(ix) FEATURE:

25 (A) NAME/KEY: exon

(B) LOCATION: 6022..6286

(ix) FEATURE:

(A) NAME/KEY: exon

30 (B) LOCATION: 6374..6570

(ix) FEATURE:

(A) NAME/KEY: exon

(B) LOCATION: 6655..7005

35

(ix) FEATURE:

(A) NAME/KEY: exon

(B) LOCATION: 7088..8032

40

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### (x1) SEQUENCE DESCRIPTION: SEQ ID NO:3:

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5	CCACTCGATT	GTCTAGGAGA	AGCCTAAGCC	GGAGTCCCAT	TCGATCACCT	AGGAAGAGTG	120
	TGAGCAGGAG	TCCAGTCCGA	TCATCTAGGA	AGAGTGTGAG	CAGAAGTCCG	GTTCGTTCAT	180
10	CCAGGAGACG	TATCAGCAGG	AGTCCAGTCC	GATCATCTAG	GAAGAGTGTG	AGCAGGAGTC	240
	CTATTCGATT	GTCCAGAAGA	AGTATCAGCA	GGAGTCCTAT	TCGATTGTCC	AGGAGAAGTA	300
	TCAGCAGGAG	TCCTGTTAGA	GGAAGAAGAA	GAATTAGCAG	AAGTCCAGTT	CCGGCAAGGA	360
15	GAAGGAGTGT	GCGGCCTAGA	тстсстсстс	CTGACCGCAG	AAGAAGTTTG	TCAAGAAGTG	420
	СТТСТССТАА	TGGGCGCATA	AGGAGAGGGA	GAGGATTTAG	CCAAAGATTC	TCATACGCCC	480
20	GTCGATACAG	AACTAGTCCA	TCTCCTGATC	GATCTCCTTA	TCGCTTTAGT	GATAGGAGTG	540
20	ACCGTGACAG	GTGAATAGCC	CACACATAAT	ATAACTCCCC	CTTTCTGTTA	CACACTCTCG	600
	TACTGAACCG	TCTTTTTTAT	AACGTCTTCT	CTGTAGATTT	AGAAGTCGCA	GAAGGTTCTC	660
25	GCCAAGTCGG	TTCAGAAGCC	CACTAAGAGG	AAGAACACCT	CCAAGGTACT	TATCCTCTTT	720
	AGTACATTGT	TTCAGCTGAT	TCTTTACATC	TAAAAGTTTC	ATGAATATGG	AACTAAAATT	780
30	GGTGATCCAA	AAGAATTATT	CTTGATTTCA	CAACTCGAAA	GTATGCTCAG	GTATAGAAGA	840
30	AGAAGCCGCT	CAGTATCGCC	TGGTCTCTGT	TATCGCAACC	GGCGGTACAG	CCGCAGCCCT	900
	ATCCGTAGCC	GATCTCCACC	TTACAGAAAG	AGAAGGTCAC	CATCCGCTAG	CCACAGCCTG	960
35	AGTCCATCGA	GGTCAAGATC	AAGATCAAAG	TCATATTCAA	AATCTCCCAT	TGGGACGGGG	1020
	AAAGCAAGAT	CAGTGTCAAG	ATCACCATCC	AAGGCAAGGT	CTCCATCGAA	GTCGGATTCG	1080
40	ACATCCTCGG	ATAATAGCCC	AGGTGGGAAA	AAGGGATTAG	TAGCCTATGA	TTAATGAATA	1140

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	ATGATTACCC	TTAAGTTAAG	TGTTTGTTCT	TTTTACTGAG	AAGAGATGGT	AAAGAGAGTA	1200
	AGTAGTTTAC	TTCTGTAAAA	CATAAGCATT	GTCTTTTGCG	TATGTTTGTT	TGATTATGCT	1260
5	CCAAGATTGT	TAAAAATTTC	TGTTGATGTT	TGCCGACATT	TTTTCTTTGT	TGCCATTTGC	1320
	CGACAAATGT	TAACTTCCAT	TATTCGTTGC	GGAGTTGGTT	TTGGTCCAAT	AATTAAACTT	1380
10	TCATAAAATT	AAGCATAACT	AAATGTGACG	TTTGTCACCA	AACTTTAGAA	CAACGACATC	1440
10	GTAATTTATT	TATTTGGATA	ATCAATATAA	TTTACGATTT	CTTCCTACAT	АТАТАТСАТА	1500
	TCACTATACC	ACCGTCATTA	TCACTATCAC	ТАААТАТААА	AATGTTAAAA	TGATTTCTTA	1560
15	ATGGAATTTT	TTTTGTTAAA	AGTTTATTGA	САСАААААТ	GAATTAAAAC	TCAGAAATCT	1620
	GTATACTGAA	TTAAAACTTG	тааататаас	AACAAAATGG	GATTAAAAAA	AGAAGTGGCA	1680
20	TCCATTTAAA	AATTATTTGC	GAATTCGCCC	GTAACTTCTT	AAGCTAACAA	TTAGAACCTA	1740
20	ATCAACACTA	GTTATTTTGA	GTCCACCGAC	AGGTGATAGC	AAATAAAAA	GAACAGGCTG	1800
	GTACCAGAGC	CAACAACAAC	GTGGCTTCTT	CTTTTTTTT	TTTAATATAA	TCAAACAATC	1860
25	ATACTTTGTC	CTATCTCTTT	CTTGCAATAA	GATTTTGCCA	CGTCACATAC	TAAGAAGCTG	1920
	GCGCGTCTAG	TGGGGAAGCC	AGAACGGCTC	ACTTTAAAAA	GTAGAGAGAT	GATAACTTGA	1980
30	GCCGAATAGA	GCCGAGCTGA	GCTAAAACGG	TGGGAGAGGA	AGAGGCTACT	ACTACCGTCA	2040
	CCATCTCCGG	TAAAATAATG	TACTTGTCAT	TTAAAAATTA	AGAAAAAACA	CATCACTCTG	2100
	CGATAAAATA	GGCAAAAGCA	GATTTGAAGA	AGAAGCAGCT	TGAGATATCA	AATAGAGAGA	2160
35	GAGAGTGACA	GAGGAGTGTG	TGAACATCCT	TTTTTAGTAG	ATTTGGGTTT	TCGAGATGCC	2220
	GTATTGAATC	GGCTACGAAT	TTCCCAATTT	TGAATTTTGT	GAATCTCTCT	CTTTCTCTGT	2280
40	GTGTCGGTGG	CTGCGATGGA	GGCCAGTGCC	GGCTTGGTTG	CTGGATCCTA	CCGGAGAAAC	2340
-							

	GAGCTCGTTC	GGATCCGACA	TGAATCTGAT	GGCGGGGTCT	GTTCATCTTC	CCTTTTTCCC	2400
	ATTTTTTTGT	TATTGTTTTT	CGTTCTTACA	ATTTTTGATG	TGTAGATCTC	ATCTAGATTT	2460
5	CTCTGTTTCT	AAATCTCGTC	TCTTTTGGAT	CCATAATTGG	ATCATTGAAA	CTCAGATTTC	2520
	GCTTCCTTTG	ACTGTGTAGT	TAGTTAGTGT	CAGTTGATCA	AGTAAGTGTC	TGAAAATGGA	2580
10	AACTTTTCTG	CTCCAATTCT	TCAAATTGTT	GTGATCTATA	TCAATTAATG	CCGCATCTGT	2640
	ТТТСТТАААА	TCTCTTATGG	AAAGTGTCGG	TGGATTTCAG	TTCGTTAACT	TTTTTAAGCT	2700
	AAAATCTTTG	ACTCTTAAAG	TTTAGCTTTA	CTTATTGAGA	TTTAGCTCAA	CTAGATCTCG	2760
15	TTAGTTCCCG	CCATGGGATA	CAGACTGTGA	CTCGCCTTAA	TTCAGATCTG	CATTGATTGT	2820
	TTTGATTTAG	ATCCTTGCTC	ATCTCTTTCT	GTAGTTTCTA	ATACTCAATG	ACTAACAATG	2880
20	ATGCAATGTT	GGTCAAAGTG	CAGACCAAAC	CTTTGAAGAA	TATGAATGGC	CAGATATGTC	2940
20	AGATCTGTGG	TGATGATGTT	GGACTCGCTG	AAACTGGAGA	TGTCTTTGTC	GCGTGTAATG	3000
	AATGTGCCTT	CCCTGTGTGT	CGGCCTTGCT	ATGAGTACGA	GAGGAAAGAT	GGAACTCAGT	3060
25	GTTGCCCTCA	ATGCAAGACT	AGATTCAGAC	GACACAGGGG	TCAGTTGTCT	ттттстттт	3120
	GTTGGCAATT	GCTATATATG	GATTTTCTCT	TTTTGTTTCT	TTGCTGTTGT	GTTGAACAAT	3180
30	TTTTTGGAAT	TTTCCAGGGA	GTCCTCGTGT	TGAAGGAGAT	GAAGATGAGG	ATGATGTTGA	3240
50	TGATATCGAG	AATGAGTTCA	ATTACGCCCA	GGGAGCTAAC	AAGGCGAGAC	ACCAACGCCA	3300
	TGGCGAAGAG	TTTTCTTCTT	CCTCTAGACA	TGAATCTCAA	CCAATTCCTC	TTCTCACCCA	3360
35	TGGCCATACG	GTAGGGACCT	ACATTTTCCC	TTTAGACTCT	AGAGTGATTT	GTATTACTCA	3420
	ATAAATCCCT	AGAGTGGTCA	TTTATTACTT	ACTATTCACG	TTAATGTTAT	ATGTGAACAA	3480
40	ATCTTAACAG	AATTTTTTC	TGATAGTACA	TGGTCATCCA	AATTAAGAAA	TAATAATAGA	3540
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	TGTTGTTAGT	TGTGTCTGTT	TTCAATAGAT	TCATGACCTT	TTTCTATACA	CAGGTTTCTG	3600
	GAGAGATTCG	CACGCCTGAT	ACACAATCTG	TGCGAACTAC	ATCAGGTCCT	TTGGGTCCTT	3660
5	CTGACAGGAA	TGCTATTTCA	TCTCCATATA	TTGATCCACG	GCAACCTGGT	ATTCATATGT	3720
	TTTTCCCTTG	TGCACGTGGT	CTTTGTTAAA	TGTGATTCCT	ATTCATTTTT	ACAACATATA	3780
10	TATTTTGTGT	ACCGTAACTG	ATAGCTCCCG	CTAAAAATTG	CAGTCCCTGT	AAGAATCGTG	3840
••	GACCCGTCAA	AAGACTTGAA	CTCTTATGGG	CTTGGTAATG	TTGACTGGAA	AGAAAGAGTT	3900
	GAAGGCTGGA	AGCTGAAGCA	GGAGAAAAT	ATGTTACAGA	TGACTGGTAA	ATACCATGAA	3960
15	GGGAAAGGAG	GAGAAATTGA	AGGGACTGGT	TCCAATGGCG	AAGAACTCCA	AATGTAAGTG	4020
	GAAATACTAG	ACCAATATCT	TTATTGTCCA	ACTCAAACAG	CTCTTGGCCG	TGATGCTAAT	4080
20	AACCACTCTT	GGTTTCTTAT	TATGTATTGA	TAGACATAAT	TAAGTATCTG	CTTTGTTACA	4140
	TTTGTTTCCT	TCCACTCAAT	TATGGTTCTC	GTACTTACAG	GGCTGATGAT	ACACGTCTTC	4200
	CTATGAGTCG	TGTGGTGCCT	ATCCCATCTT	CTCGCCTAAC	CCCTTATCGG	GTTGTGATTA	4260
25	TTCTCCGGCT	TATCATCTTG	TGTTTCTTCT	TGCAATATCG	TACAACTCAC	CCTGTGAAAA	4320
	ATGCATATCC	TTTGTGGTTG	ACCTCGGTTA	TCTGTGAGAT	CTGGTTTGCA	TTTTCTTGGC	4380
30	TTCTTGATCA	GTTTCCCAAA	TGGTACCCCA	TTAACAGGGA	GACTTATCTT	GACCGTCTCG	4440
	CTATAAGGTT	GGTCTTTAAG	TTTATACATC	CCCTACTCTC	ATCTCTCTTT	TATGTATTAA	4500
	CTTGATATCT	TCTATCACAG	TTTTCGATAG	TTGACTTTTT	CCCCCTGTAA	ATTTAATTTA	4560
35	AATTTAGACA	ATGGTGCATC	TGAATTTTGA	TTATGATATA	TCTTAAGAAG	ATTATGATTG	4620
	TAAATCTTGA	AATTTAGTAG	AAAACCATCT	GCAATCTACT	GACCATGTGA	AGTTTCCGAC	4680
40	TAGACTATGA	TAGAAGCATG	CCAAGTGGAG	TGTTTATTAA	GATAGAGCTT	AGCTATTATA	4740

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	CTGATTTTAT	ATGTGTTTTG	ATTTTTTGGT	TTCTTATTGT	AGATATGATC	GAGACGGTGA	4800
	ACCATCACAG	CTCGTTCCTG	TTGATGTGTT	TGTTAGTACA	GTGGACCCAT	TGAAAGAGCC	4860
5	TCCCCTTGTT	ACAGCAAACA	CAGTTCTCTC	GATTCTTTCT	GTGGACTACC	CGGTAGATAA	4920
	AGTAGCCTGT	TATGTTTCAG	ATGATGGTTC	AGCTATGCTT	ACCTTTGAAT	CCCTTTCTGA	4980
10	AACCGCTGAG	TTTGCAAAGA	AATGGGTACC	ATTTTGCAAG	AAATTCAACA	TTGAACCTAG	5040
10	GGCCCCTGAA	TTCTATTTTG	CCCAGAAGAT	AGATTACTTG	AAGGACAAGA	TCCAACCGTC	5100
	TTTTGTTAAA	GAGCGACGAG	CTATGAAGGT	CATTTGAAAA	GTCCACCTGC	TTCTCATCCA	5160
15	TACGGCAAAG	AGATTGACTG	ACTTTTTCTT	TGGTTTGTAT	TGACAGAGAG	AGTATGAAGA	5220
	GTTTAAAGTG	AGGATAAATG	CTCTTGTTGC	CAAAGCACAG	AAAATCCCTG	AAGAAGGCTG	5280
20	GACAATGCAG	GATGGTACTC	CCTGGCCTGG	TAACAACACT	AGAGATCATC	CTGGAATGAT	5340
20	ACAGGTACAG	TGTGGCAATC	CCTTGATTGT	GACAGAGAGG	ATAACGTAAA	GGAAACATGT	5400
	TTACATCGTT	TTGTTTCAAT	TTCAGGTGTT	CTTAGGCCAT	AGTGGGGGTC	TGGATACCGA	5460
25	TGGAAATGAG	CTGCCTAGAC	TCATCTATGT	TTCTCGTGAA	AAGCGGCCTG	GATTTCAACA	5520
	CCACAAAAAG	GCTGGAGCTA	TGAATGCATT	GGTTTGTTAA	CTTTCAGAAT	CCTATTGTGT	5580
30	CCTCTATTTT	ATTCTCTTGT	TCACTGCCTA	AGAAACGTTC	TTCTTGTGTA	GCCGTTGCTT	5640
30	CACATTCTTT	TTTTTCTAGG	CTATGTGTTC	TCTCCTAATT	TAGTATCTCT	TTACTTTGAC	5700
	AGATCCGTGT	ATCTGCTGTT	CTTACCAATG	GAGCATATCT	TTTGAACGTG	GATTGTGATC	5760
35	ATTACTTTAA	TAACAGTAAG	GCTATTAAAG	AAGCTATGTG	TTTCATGATG	GACCCGGCTA	5820
	TTGGAAAGAA	GTGCTGCTAT	GTCCAGTTCC	CTCAACGTTT	TGACGGTATT	GATTTGCACG	5880
40	ATCGATATGC	CAACAGGAAT	ATAGTCTTTT	TCGATGTGAG	TATCACTTCC	CCATTGTCTT	5940

	TTGTTTCTCT	TTTGTTCATA	TTTTGGTTGG	ATTTACTCGT	TTCTGCTATG	GCCTGACTTG	6000
	GATATTTGTT	CTCTTGGGCA	GATTAACATG	AAGGGGTTGG	ATGGTATCCA	GGGTCCAGTA	6060
5	TATGTGGGTA	CTGGTTGTTG	TTTTAATAGG	CAGGCTCTAT	ATGGGTATGA	TCCTGTTTTG	6120
	ACGGAAGAAG	ATTTAGAACC	AAATATTATT	GTCAAGAGCT	GTTGCGGGTC	AAGGAAGAAA	6180
10	GGTAAAAGTA	GCAAGAAGTA	TAACTACGAA	AAGAGGAGAG	GCATCAACAG	AAGTGACTCC	6240
	AATGCTCCAC	TTTTCAATAT	GGAGGACATC	GATGAGGGTT	TTGAAGGTTT	GATTGAGCTG	6300
	ATTGTGTAAT	AACATCACTT	CTTTATGTAA	TGATTTATGT	GATGGTGAAA	TCTTACAATC	6360
15	CTTGTTTATG	CAGGTTATGA	TGATGAGAGG	TCTATTCTAA	TGTCCCAGAG	GAGTGTAGAG	6420
	AAGCGTTTTG	GTCAGTCGCC	GGTATTTATT	GCGGCAACCT	TCATGGAACA	AGGCGGCATT	6480
20	CCACCAACAA	CCAATCCCGC	TACTCTTCTG	AAGGAGGCTA	TTCATGTTAT	AAGCTGTGGT	6540
20	TACGAAGACA	AGACTGAATG	GGGCAAAGAG	GTCAGTTTTC	AAATGCAGCT	ACAGAATCTT	6600
	CTTATGTTCT	CTTTCTTACC	TGTTTGATGA	CATCTTATTT	GGCACTTTTG	TTAGATTGGT	6660
25	TGGATCTATG	GTTCCGTGAC	GGAAGATATT	CTTACTGGGT	TCAAGATGCA	TGCCCGGGGT	6720
	TGGATATCGA	TCTACTGCAA	TCCTCCACGC	CCTGCGTTCA	AGGGATCTGC	ACCAATCAAT	6780
30	CTTTCTGATC	GTTTGAACCA	AGTTCTTCGA	TGGGCTTTGG	GATCTATCGA	GATTCTTCTT	6840
50	AGCAGACATT	GTCCTATCTG	GTATGGTTAC	CATGGAAGGT	TGAGACTTTT	GGAGAGGATC	6900
	GCTTATATCA	ACACCATCGT	CTATCCTATT	ACATCCATCC	CTCTTATTGC	GTATTGTATT	6960
35	CTTCCCGCTT	TTTGTCTCAT	CACCGACAGA	TTCATCATAC	CCGAGGTTTG	TAAAACTGAC	7020
	CACACTGCTA	TTTACTATTT	GAATCCCATT	TTGTGAATGC	ATTTTTTGT	CATCATCATT	7080
40	GTTGCAGATA	AGCAACTACG	CGAGTATTTG	GTTCATTCTA	CTCTTCATCT	CAATTGCTGT	7140

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	GACTGGAATC	CTGGAGCTGA	GATGGAGCGG	TGTGAGCATT	GAGGATTGGT	GGAGGAACGA	7200
	GCAGTTCTGG	GTCATTGGTG	GCACATCCGC	CCATCTTTTT	GCTGTCTTCC	AAGGTCTACT	7260
5	TAAGGTTCTT	GCTGGTATCG	ACACCAACTT	CACCGTTACA	TCTAAAGCCA	CAGACGAAGA	7320
	TGGGGATTTT	GCAGAACTCT	ACATCTTCAA	ATGGACAGCT	CTTCTCATTC	CACCAACCAC	7380
10	CGTCCTACTT	GTGAACCTCA	TAGGCATTGT	GGCTGGTGTC	TCTTATGCTG	TAAACAGTGG	7440
	CTACCAGTCG	TGGGGTCCGC	TTTTCGGGAA	GCTCTTCTTC	GCCTTATGGG	TTATTGCCCA	7500
	TCTCTACCCT	TTCTTGAAAG	GTCTGTTGGG	AAGACAAAAC	CGAACACCAA	CCATCGTCAT	7560
15	TGTCTGGTCT	GTTCTTCTCG	CCTCCATCTT	CTCGTTGCTT	TGGGTCAGGA	TCAATCCCTT	7620
	TGTGGACGCC	AATCCCAATG	CCAACAACTT	CAATGGCAAA	GGAGGTGTCT	TTTAGACCCT	7680
20	ATTTATATAC	TTGTGTGTGC	АТАТАТСААА	AACGCGCAAT	GGGAATTCCA	AATCATCTAA	7740
	ACCCATCAAA	CCCCAGTGAA	CCGGGCAGTT	AAGGTGATTC	CATGTCCAAG	ATTAGCTTTC	7800
	TCCGAGTAGC	CAGAGAAGGT	GAAATTGTTC	GTAACACTAT	TGTAATGATT	TTCCAGTGGG	7860
25	GAAGAAGATG	TGGACCCAAA	TGATACATAG	TCTACAAAA	GAATTTGTTA	TTCTTTCTTA	7920
	TATTTATTTT	ATTTAAAGCT	TGTTAGACTC	ACACTTATGT	AATGTTGGAA	CTTGTTGTCC	7980
30	TAAAAAGGGA	TTGGAGTTTT	СТТТТТАТСТ	AAGAATCTGA	AGTTTATATG	CTAAGCTTTT	8040
	CACTTTACTA	CAAAAAGTTT	ATGGATATGA	TGGTGTACGT	CAATTGTTGG	TGCAAGTGTT	8100
	GATGTCTTCG	GGTGAACTCG	CCCTCTTGTT	TTGTCTCACC	CATCAGTACA	AATAGAATGA	8160
35	CATTTATTTT	TTTGAACTTT	TAACGAAATC	TTTGTCATTA	TGGGACTTGA	TCAGTAAAGT	8220
	TACATATTTG	AAGAGATATT	GTGTAAACTC	TTATTTGAAT	CAGAATCAGA	TCAATCAAAA	8280
40	ATTGAAAACG	TAAAGTTCAA	ACAAAAAGGT	AGAGTGAATC	TTTTAATCCC	CCCTCAATAC	8340
. •							

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8400 TAATTTGTGA AATCTCAAGT GGTGTAAAAT GAACCCAATT AGTATCCACA ATGTGTTTCT 8411 CTGATCAATC C 5 (2) INFORMATION FOR SEQ ID NO:4: (i) SEQUENCE CHARACTERISTICS: 10 (A) LENGTH: 5009 base pairs (B) TYPE: nucleic acid (C) STRANDEDNESS: single (D) TOPOLOGY: linear 15 (ii) MOLECULE TYPE: DNA (genomic) (iii) HYPOTHETICAL: NO (iv) ANTI-SENSE: NO 20 (vi) ORIGINAL SOURCE: (A) ORGANISM: Arabidopsis thaliana (B) STRAIN: Columbia 25 (vii) IMMEDIATE SOURCE: (B) CLONE: 12C4 (ix) FEATURE: (A) NAME/KEY: exon 30 (B) LOCATION: 863..943 (ix) FEATURE: (A) NAME/KEY: exon (B) LOCATION: 1454..1840 35 (ix) FEATURE: (A) NAME/KEY: exon (B) LOCATION: 1923..2025

40

(ix) FEATURE:

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(A) NAME/KEY: exon

		(B)	LOCATION:	21222311				
	(ix)	FEAT	URE:					
5		(A)	NAME/KEY:	exon				
		(B)	LOCATION:	24212687				
	(ix)	FEAT	URE:					
		(A)	NAME/KEY:	exon				
10		(B)	LOCATION:	27763121				
	(ix)	FEAT	URE:					
		(A)	NAME/KEY:	exon				
15		(B)	LOCATION:	32203357				
1,5	(ix)	FEAT	URE:					
		(A)	NAME/KEY:	exon				
		(B)	LOCATION:	35073623				
20	(ix)	FEAT	URE:					
		(A)	NAME/KEY:	exon				
		(B)	LOCATION:	37233935				
	(ix)	FEAT	URE:					
25		(A)	NAME/KEY:	exon				
		(B)	LOCATION:	40274297	,			
	(ix)	FEAT	URE:					
		(A)	NAME/KEY:	exon				
30		(B)	LOCATION:	43804576	i			
	(xi)	SEQU	ENCE DESCR	IPTION: SEC	) ID NO:4:			
35	AAGGAATA	AA TA	GATAGGGG T	TTAATGGGA G	ACAATCAAT	CTTCAGGGGT	TTTCTGGAAN	60
	AACGGCGG	GG TA		ACATCAATC G	GACCCGATC	ACGAGGACCC	GGATCCGNAT	120
40	CGATAAAC	AG NG	TAGCTTTC A	атассссат т	TTCCCAGAA	ACACCTCTCA	AAAATTTTTT	180

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	CAAGAACTNG	TATAAATATC	TCAGTTTCGT	TCACGCAGGT	CTTTNTTATT	TTGGNAANTC	240
	TNTNTTCATN	GTTCACCAAC	TCCCTCTTGA	AGGTGGGACA	GAGTCCAGCT	CCACCACCAC	300
5	CATAGCCATC	GCGTCGTTTT	CTCCGGGACC	CACTTATTTC	GTGACGTTTC	TCTCTTTGTA	360
	TATACATACA	ATTGTTTTCA	GTCTCAATTT	GCTGTCCACA	TTTTAACACA	ACTCTATCTC	420
10	AGGGGTGGTG	TCTGAATCTC	GTCTCTCTCA	TTCCTATTTA	TCCCAATCTA	ATCTATCACA	480
••	AACCCTTCCA	CATTGCTTTT	GTCAGTCTGT	AAAATTCTCT	TTGAATCAGT	GAATCACTCA	540
	CTTAAATCCA	AAACAGTTTT	TTTTTCTTTC	TTTCTTTATT	TGCTTGTTGT	GGAATCAATA	600
15	GCTGTCTCCG	GGAAAATTCG	TTTTTTTCT	CCTTCGGGAT	CTCTTTTTT	TTTTTTTGG	660
	TTTTATTTAA	TAATTATCCC	CGAGCCAACA	TTTATTGTCG	ATTCGGTTTA	TTTCGTCTCC	720
20	TTCGTCTTCC	ACTCTTACTA	GTGCATGCTC	TGAATCTGTA	TGTAATGGGA	GTTCAACAGT	780
20	CTGGATCCAT	TATCCTAGCC	GGGTCGGGTC	AAGGTCTTTG	AGTAAGAGAG	ACAATTCGTT	840
	TTGATTCGGT	GTAGAAGACA	TCATGAATAC	TGGTGGTCGG	CTCATTGCTG	GCTCTCACAA	900
25	CAGAAACGAA	TTCGTTCTCA	TTAACGCCGA	TGAGAGTGCC	AGAGTAAGAA	TAACTTTTGT	960
	ANGAATTTGT	GACGGAAAAA	AGTTTAATTT	TTTCTCTTTC	TTGGGGATCT	AGATTATGAG	1020
30	AATCTAGATG	GAATATTTTG	ATCTGAAATT	GGAAGTTTCT	AGGGAGTAAT	GCCGCAACCC	1080
	ACATGTTCTG	TTTTTTCTTT	TTTCTTTCT	TCAAGTAGTG	TTGCATGATT	CATACGTGTC	1140
	GGCAGAGATG	TCCTGAGAAC	CGAATTCAAT	GTTGTAGCAG	TAGCAATAAG	TTCAAAGAAA	1200
35	GTCCATTTT	TTATATTACT	AATTCTGTTC	TIGGTTTATT	TGAGCTGGTC	TTTATTGCAT	1260
	TTCACCTGGA	TTCAGATACT	AATAACTGTC	TCAATTATGT	AAAAATGACA	ACTTTATGAA	1320
40	ATTCAGTTTC	ACAATTATGT	AATTCATAAT	CGATGAATGT	TTTTCTTGAG	TCTTTATCAT	1380

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	CTTTAGGATT	TGATTAAGAT	GCAATTTGAT	GAAAATACTA	AAAAGACTCA	TGTGTTCTCA	1440
	TTTCTCTATG	TAGATACGAT	CAGTACAAGA	ACTGAGTGGG	CAAACATGTC	AAATCTGTGG	1500
5	AGATGAAATC	GAATTAACGG	TTAGCAGTGA	GCTCTTTGTT	GCTTGCAACG	AATGCGCATT	1560
	CCCGGTTTGT	AGACCATGCT	ATGAGTATGA	ACGTAGAGAA	GGAAATCAAG	CTTGTCCTCA	1620
10	GTGCAAAACT	CGATACAAAA	GGATTAAAGG	TAGTCCACGG	GTTGATGGAG	ATGATGAAGA	1680
	AGAAGAAGAC	ATTGATGATC	TTGAGTATGA	GTTTGATCAT	GGGATGGACC	CTGAACATGC	1740
	CGCTGAAGCC	GCACTCTCTT	CACGCCTTAA	CACCGGTCGT	GGTGGATTGG	ATTCAGCTCC	1800
15	ACCTGGCTCT	CAGATTCCTC	TTTTGACTTA	TTGTGATGAA	GTGAGGAATC	CAAATTGTTT	1860
	GTTTTCTCTG	ACAATGTTGT	TGCTTAGATG	ATTCTTTTTC	TTATTAGTCT	ATGTGTTTTC	1920
20	AGGATGCTGA	TATGTATTCT	GATCGTCATG	CTCTTATCGT	GCCTCCTTCA	ACGGGATATG	1980
	GGAATCGCGT	CTATCCTGCA	CCGTTTACAG	ATTCTTCTGC	ACCTCGTATG	TGTTTACTTT	2040
	TATGATTCCT	ACAATTTTTC	TTCTTATATG	ATTTGGTCAC	CTTCTAATGA	GTTATGAAAT	2100
25	GGTTTTGTTT	GTTGTTTTCA	GCACAGGCGA	GATCAATGGT	TCCTCAGAAA	GATATTGCGG	2160
	AATATGGTTA	TGGAAGTGTT	GCTTGGAAGG	ACCGTATGGA	AGTTTGGAAG	AGACGACAAG	2220
30	GCGAAAAGCT	TCAAGTCATT	AAGCATGAAG	GAGGAAACAA	TGGTCGAGGT	TCCAATGATG	2280
50	ACGACGAACT	AGATGATCCT	GACATGCCTA	TGTAAGTTGT	TAAAATCTAA	CAAAAGTTCA	2340
	GATGAAATGA	TGCTCTGAAA	TTTTGTGTTC	AATGGNTTTG	TTTTCTTATT	GTTGTTTAAA	2400
35	CATTTTTCGT	GCTAATTCAG	GATGGATGAA	GGAAGACAAC	CTCTCTCAAG	AAAGCTACCT	2460
	ATTCGTTCAA	GCAGAATAAA	TCCTTACAGG	ATGTTAATTC	тстстсссст	CGCGATTCTT	2520
40		TTCATTATAG	AATTCTCCAT	CCAGTCAATG	ATGCATATGG	ATTATGGTTA	2580

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ACGTCAGTTA	TATGCGAGAT	ATGGTTTGCA	GTGTCTTGGA	TTCTTGATCA	ATTCCCCAAA	2640
TGGTATCCTA	TAGAACGTGA	AACATACCTC	GATAGACTCT	CTCTCAGGTA	ACATAAACCC	2700
TGAAAAGTTC	TTGTCTGCAA	ATATTCATTT	TTTACATTCC	CAAAAATTTT	TGAAACTCTA	2760
TTTTTCTTAC	ATAAGGTACG	AGAAGGAAGG	AAAACCGTCA	GGATTAGCAC	CTGTTGATGT	2820
TTTTGTTAGT	ACAGTGGATC	CGTTGAAAGA	GCCACCCTTG	ATTACAGCAA	ACACAGTTCT	2880
TTCCATTCTA	GCAGTTGATT	ATCCTGTGGA	TAAGGTTGCG	TGTTATGTAT	CAAACAATGG	2940
TGCAGCTATG	CTTACATTTG	AAGCTCTCTC	TGATACAGCT	GAGTTTGCTA	GAAAATGGGT	3000
TCCTTTTTGT	AAGAAGTTTA	ATATCGAGCC	ACGAGCTCCT	GAGTGGTATT	TTTCTCAGAA	3060
GATGGATTAC	CTGAAGAACA	AAGTTCATCC	TGCTTTTGTC	AGGGAACGTC	GTGCTATGAA	3120
GGTTTTCTTT	GCTGCTTTTT	CTCTTTCTGA	GTATATCCTA	TCATAAAAGT	GTTGTTTCAA	3180
GAATCTGATT	TACGTTTTTT	GCTTGTTTGT	TTGTTGCAGA	GAGATTATGA	GGAGTTTAAA	3240
GTGAAGATAA	ATGCACTGGT	TGCTACTGCA	CAGAAAGTGC	CTGAGGAAGG	TTGGACTATG	3300
CAAGATGGAA	CTCCTTGGCC	TGGAAACAAC	GTCCGTGACC	ATCCTGGAAT	GATTCAGGTA	3360
ATGATGAGTT	TGATTGAATA	GGCAAAAAA	AAGCGGTTTT	TGTCCTCTTC	ACTTTGTTTC	3420
CCTGGATCTG	TTAAATTGGA	ATGAGCACTC	TACTTCTCAA	тататсттса	GACCGAAGCC	3480
TTTTTAAGAG	ATTTTGTAAA	TGACAGGTGT	TCTTGGGTCA	TAGTGGAGTT	CGTGATACGG	3540
ATGGTAATGA	GTTACCACGT	CTAGTGTATG	TTTCTCGTGA	GAAGCGGCCT	GGATTTGATC	3600
ACCACAAGAA	AGCTGGAGCT	ATGAATTCCT	TGGTAAGTAT	AATGTGTTTC	TTTATTTATG	3660
AATCTCTCTT	TTCGGAGCCC	TGACTTCTCA	TAAACTAAAA	CTCATCTTAC	TTCTTCTTGA	3720
AGATCCGAGT	CTCTGCTGTT	CTATCAAACG	CTCCTTACCT	TCTTAATGTC	GATTGTGATC	3780
	TGGTATCCTA TGAAAAGTTC TTTTTCTTAC TTTTGTTAGT TTCCATTCTA TGCAGCTATG TCCTTTTTGT GAATCTGATT GAATCTGATT GTGAAGATAA CAAGATGAA ATGATGAGT TCCTGGATCTG TTTTTAAGAG ATGGTAATGA ACCACAAGAA AATCTCTCTT	TGGTATCCTA TAGAACGTGA TGAAAAGTTC TTGTCTGCAA TTTTTCTTAC ATAAGGTACG TTTTGTTAGT ACAGTGGATC TTCCATTCTA GCAGTTGATT TGCAGCTATG CTTACATTTG AAGAAGATTA GATGGATTAC CTGAAGAACA GGTTTTCTTT GCTGCTTTTT GAATCTGATT TACGTTTTTT GTGAAGATAA ATGCACTGGT CAAGATGGAA CTCCTTGGCC ATGATGAGTT TGATTGAATA CCTGGATCTG TTAAATTGGA TTTTTAAGAG ATTTTGTAAA ATGGTAATGA GTTACCACGT ACCACAAGAA AGCTGGAGCCC AATCTCTCTT TTCGGAGCCC	TGGTATCCTA TAGAACGTGA AACATACCTC TGAAAAGTTC TTGTCTGCAA ATATTCATTT TTTTTCTTAC ATAAGGTACG AGAAGGAAGG TTTTGTTAGT ACAGTGGATC CGTTGAAAGA TCCATTCTA GCAGTTGATT ATCCTGTGGA TGCAGCTATG CTTACATTTG AAGCTCTCTC GATGGATTAC CTGAAGAACA AAGTTCATCC GGTTTTCTTT GCTGCTTTTT CTCTTTCTGA GAATCTGATT TACGTTTTTT GCTTGTTGT GTGAAGATAA ATGCACTGGT TGCTACTGCA CAAGATGGAA CTCCTTGGCC TGGAAACAAC ATGATGAGT TGATTGAATA GGCAAAAAAA CCTGGATCTG TTAAATTGGA ATGAGCACTC TTTTTAAGAG ATTTTGTAAA TGACAGGTGT ATGGTAATGA GTTACCACGT CTAGTGTATG ACCACAAGAA AGCTGGAGCC TGACTTCTCA	TGGTATCCTA TAGAACGTGA AACATACCTC GATAGACTCT TGAAAAGTTC TTGTCTGCAA ATATTCATTT TTTACATTCC TTTTTCTTAC ATAAGGTACG AGAAGGAAGG AAAACCGTCA TTTTGTTAGT ACAGTGGATC CGTTGAAAGA GCCACCCTTG TCCATTCTA GCAGTTGATT ATCCTGTGGA TAAGGTTGCG TGCAGCTATG CTTACATTTG AAGCTCTCC TGATACAGCT TCCTTTTTGT AAGAAGTTTA ATATCGAGCC ACGAGCTCCT GATGGATTAC CTGAAGAACA AAGTTCATCC TGCTTTTGTC GGTTTTCTTT GCTGCTTTTT CTCTTTCTGA GTATATCCTA GAATCTGATT TACGTTTTTT GCTTGTTTGT TTGTTGCAGA GTGAAGATAA ATGCACTGGT TGCTACTGCA CAGAAAGTGC CAAGATGGAA CTCCTTGGCC TGGAAACAAC GTCCGTGACC ATGATGAGTT TGATTGAATA GGCAAAAAAA AAGCGGTTTT CCTGGATCTG TTAAATTGGA ATGAGCACTC TACTTCTCAA TTTTTAAGAG ATTTTGTAAA TGACAGGTGT TCTTGGGTCA ATGGTAATGA GTTACCACGT CTAGTGTATG TTTCTCGTGA ACCACAAGAA AGCTGGAGCT ATGAATTCCT TGGTAAGTAT AATCTCTCTT TTCGGAGCCC TGACTTCTCA TAAACTAAAA	TGGTATCCTA TAGAACGTGA AACATACCTC GATAGACTCT CTCTCAGGTA TGAAAAGTTC TTGTCTGCAA ATATTCATTT TTTACATTCC CAAAAATTTT TTTTTCTTAC ATAAGGTACG AGAAGGAAGG AAAACCGTCA GGATTAGCAC TTTTGTTAGT ACAGTGGATC CGTTGAAAGA GCCACCCTTG ATTACAGCAA TTCCATTCTA GCAGTTGATT ATCCTGTGGA TAAGGTTGCG TGTTATGTAT TGCAGCTATG CTTACATTTG AAGCTCTCC TGATACAGCT GAGTGTGCTA TCCTTTTTGT AAGAAGTTTA ATATCGAGCC ACGAGCTCCT GAGTGGTATT GATGGATTAC CTGAAGAACA AAGTTCATCC TGCTTTTGTC AGGGAACGTC GGTTTTCTTT GCTGCTTTTT CTCTTTCTGA GTATATCCTA TCATAAAAGT GAATCTGATT TACGTTTTTT GCTTGTTTGT TTGTTGCAGA GAGATTATGA GTGAAGATAA ATGCACTGGT TGCTACTGCA CAGAAAGTGC CTGAGGAAGG CAAGATGGAA CTCCTTGGCC TGGAAACAAC GTCCGTGACC ATCCTGGAAT ATGATGAGTT TGATTGAATA GGCAAAAAAA AAGCGGTTTT TGTCCTCTTC CCTGGATCTG TTAAATTGGA ATGAGCACTC TACTTCCAA TATATCTTCA TTTTTAAGAG ATTTTGTAAA TGACAGGTGT TCTTGGGTCA TAGTGGAGTT ATGGTAATGA GTTACCACGT CTAGTGTATG TTTCTCGTGA GAAGCGGCCT ACCACAAGAA AGCTGGAGCT ATGAATTCCT TGGTAAGTAT AATGTGTTTC AATCTCTCTT TTCGGAGCCC TGACTTCTCA TAAACTAAAA CTCATCTTAC	TOGTATICA TAGACCIGA AACATACCTC GATAGACTCT CTCTCAGGTA ACATAAACCC TGAAAAGTTC TTGTCTGCAA ATATTCATTT TITACATTCC CAAAAATTTT TGAAACTCTA TTTTTCTTAC ATAAGGTACG AGAAGGAAGG AAAACCGTCA GGATTAGCAC CTGTTGATGT TTTTGTTAGT ACAGTGGATC CGTTGAAAGA GCCACCCTTG ATTACAGCAA ACACAGTTCT TTCCATTCTA GCAGTTGATT ATCCTGTGGA TAAGGTTGCG TGTTATGTAT CAAACAATGG TGCAGCTATG CTTACATTTG AAGCTCTCC TGATACAGCT GAGTTGGTAT TTTCTCAGAA GATGGATTAC CTGAAGAACA AAGTTCATCC TGCTTTTGTC AGGGAACGTC GTGCTATGAA GGTTTTCTTT GCTGCTTTTT CTCTTTCTGA GTATATCCTA TCATAAAAGT GTTGTTTCAA GGATCTGATT TACGTTTTTT GCTTGTTTGT TTGTTCAGGA GAGAGTATAGA GGAGTTTAAA GTGAAGATAA ATGCACTGGT TGCTACTGCA CAGAAAGTGC CTGAGGAAGG TTGGACTATG CAAGATGGAA CTCCTTGGCC TGGAAACAAC GTCCGTGACC ATCCTGGAAT GATTCAGGTA ATGATGAGTT TGATTGAATA GGCAAAAAAA AAGCGGTTTT TGTCCTCTTC ACTTTGTTC CCTGGATCTG TTAAATTGGA ATGAGCACTC TACTTCCTAA TATATCTTCA GACCGAAGCC TTTTTAAGAG ATTTTGTAAA TGACAGGTGT TCTTCGCTGA TAGTGGAGTT COTGATACCG ATGGTAATGA GTTACCACGT CTAGTGTATG TTCTCCTGTA TAGTGGAGTT CTTGATTC ACCACAAGAA AGCTGGAGCT ATGAATTCCT TGGTAAGTAT AATGTGTTTC TTTATTTTTC AATCCTCTT TTCGGAGCCC TGACTTCTCA TAAACTAAAA CTCATCTTAC TTCTTCTTGA AAGTCCCACAGCT CTGCTGGTT CTATCAACCG CTCCTTACCT TCTTAATTCC GATTGTGTC AGGTCCGAGT CTCTCCTTTC TTATCTCCA TAAACTAAAA CTCATCTTAC TTCTTCTTGA AGGTCCGAGT CTCTCCTTTC TTATCTCTAC TAAACTAAAA CTCATCTTAC TTCTTCTTGA AGGTCCGAGT CTCTCCTTTC TTATCTCTAC TCCTTTAATTCT GATTGTGTCC AGGTCCGAGT CTCTCCTTTC TTATCTACCT TCTTAATTCT GATTGTGTC

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	ACTACATCAA	CAACAGCAAA	GCAATTAGAG	AATCTATGTG	TTTCATGATG	GACCCGCAAT	3840
	CGGGAAAGAA	AGTTTGTTAT	GTTCAGTTTC	CGCAGAGATT	TGATGGGATT	GATAGACATG	3900
5	ATAGATACTC	AAACCGTAAC	GTTGTGTTCT	TTGATGTATG	TGTCCTTATC	TCTTTTGCTT	3960
	TGTTTCTGTT	TATGTTTAG	TGCTTTTCCT	CTTTTCTCAT	TTGATATTGT	TTTGGTGTGG	4020
10	AAACAGATTA	ACATGAAAGG	TCTTGATGGG	ATACAAGGAC	CGATATATGT	CGGGACAGGT	4080
••	TGTGTGTTTA	GAAAACAGGC	TCTTTATGGT	TTTGATGCAC	CAAAGAAGAA	GAAACCACCA	4140
	GGCAAAACCT	GTAACTGTTG	GCCTAAATGG	TGTTGTTTGT	GTTGTGGGTT	GAGAAAGAAG	4200
15	AGTAAAACGA	AAGCCAAAGA	TAAGAAAACT	AACACTAAAG	AGACTTCAAA	GCAGATTCAT	4260
	GCGCTAGAGA	ATGTCGACGA	AGGTGTTATC	GTCCCAGGTA	AAAAAAGAAG	GAAAAAAAA	4320
20	ACATTTCTTA	TTTGGTTTCT	GTCTTGTTGA	AAGTCTAAGT	AGATCCTTTT	GATTGTTAGT	4380
	GTCAAATGTT	GAGAAGAGAT	CTGAAGCAAC	ACAATTGAAA	TTGGAGAAGA	AGTTTGGACA	4440
	ATCTCCGGTT	TTCGTTGCCT	CTGCTGTTCT	ACAGAACGGT	GGAGTTCCCC	GTAACGCAAG	4500
25	CCCCGCATGT	TTGTTAAGAG	AAGCCATTCA	AGTTATTAGC	TGCGGGTACG	AAGATAAAAC	4560
	CGAATGGGGA	AAAGAGGTAG	AAAACATTAC	AAAGTTTTTC	AACTTCTGAA	AACTAGAAAA	4620
30	GTTCTTGTGA	TCTCATTCTT	GCTGATAATC	ACACGCAGAT	CGGGTGGATT	TATGGATCGG	4680
	TGACTGAAGA	TATCCTGACG	GGTTTCAAGA	TGCATTGCCA	TGGATGGAGA	TCTGTGTACT	4740
	GTATGCCTAA	GCGTGCAGCT	TTTAAAGGAT	CTGCTCCTAT	TAACTTGTCA	GATCGTCTTC	4800
35	ATCAAGTTCT	ACGTTGGGCT	CTTGGCTCTG	TAGAGATTTT	CTTGAGCAGA	CATTGTCCGA	4860
	TATGGTATGG	TTATGGTGGT	GGTTTAAAAT	GGTTGGAGAG	ATTCTCTTAC	ATCAACTCTG	4920
40	TCGTCTATCC	TTGGACTTCA	CTTCCATTGA	TCGTCTATTG	TTCTCTCCCC	GCGGTTTGTT	4980
. •							

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TACTCACAGG AAAATTCATC GTCCCTGAG

5009

96

30

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5 (2) INFORMATION FOR SEQ ID NO:5:
        (i) SEQUENCE CHARACTERISTICS:
             (A) LENGTH: 3603 base pairs
             (B) TYPE: nucleic acid
10
             (C) STRANDEDNESS: single
             (D) TOPOLOGY: linear
       (ii) MOLECULE TYPE: cDNA
15
     (iii) HYPOTHETICAL: NO
       (vi) ORIGINAL SOURCE:
             (A) ORGANISM: Arabidopsis thaliana
             (B) STRAIN: Columbia
20
     (vii) IMMEDIATE SOURCE:
             (B) CLONE: RSW1 cDNA
       (ix) FEATURE:
25
             (A) NAME/KEY: CDS
             (B) LOCATION: 1..3243
       (xi) SEQUENCE DESCRIPTION: SEQ ID NO:5:
30
   ATG GAG GCC AGT GCC GGC TTG GTT GCT GGA TCC TAC CGG AGA AAC GAG
                                                                         48
   Met Glu Ala Ser Ala Gly Leu Val Ala Gly Ser Tyr Arg Arg Asn Glu
     1
                                                            15
```

35 CTC GTT CGG ATC CGA CAT GAA TCT GAT GGC GGG ACC AAA CCT TTG AAG

20

Leu Val Arg Ile Arg His Glu Ser Asp Gly Gly Thr Lys Pro Leu Lys

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	AAT	ATG	AAT	GGC	CAG	ATA	TGT	CAG	ATC	TGT	GGT	GAT	GAT	GTT	GGA	CTC	144
	Asn	Met	Asn	Gly	Gln	Ile	Cys	Gln	Ile	Cys	Gly	Asp	Asp	Val	Gly	Leu	
			35					40					45				
_																	
5												GAA					192
	Ala	Glu	Thr	Gly	qeA	Val	Phe	Val	Ala	Cys	Asn	Glu	Cys	Ala	Phe	Pro	
		50					55					60					
10												GAT					240
IU		Cys	Arg	Pro	Суѕ		Glu	Tyr	Glu	Arg		Asp	Gly	Thr	Gln	_	
	65					70					75					80	
	<b>6</b> 00		<b>CNN</b>	<b>800</b>		N CITT	202	mmo		<b>~~</b>	010		999				
												AGG					288
15	cys	PIO	GIN	cys	85	inr	Arg	Pne	Arg	_	HIS	Arg	GIÀ	ser		Arg	
1 3					03					90					95		
	GTT	AAD	GGA	GAT	GAA	ТАЭ	GAG	CAT	CAT	CTT	СУТ	GAT	אדר	CAG	ከ ከ ጥ	CAG	336
												Asp					330
			1	100					105				•••	110	21011	010	
20																	
	TTC	AAT	TAC	GCC	CAG	GGA	GCT	AAC	AAG	GCG	AGA	CAC	CAA	CGC	CAT	GGC	384
	Phe	Asn	Tyr	Ala	Gln	Gly	Ala	Asn	Lys	Ala	Arg	His	Gln	Arg	His	Gly	
			115					120					125				
25	GAA	GAG	TTT	TCT	TCT	TCC	TCT	AGA	CAT	GAA	TCT	CAA	CCA	ATT	CCT	CTT	432
	Glu	Glu	Phe	Ser	Ser	Ser	Ser	Arg	His	Glu	Ser	Gln	Pro	Ile	Pro	Leu	
		130					135					140					
	CTC	ACC	CAT	GGC	CAT	ACG	GTT	TCT	GGA	GAG	ATT	CGC	ACG	CCT	GAT	ACA	480
30	Leu	Thr	His	Gly	His	Thr	Val	Ser	Gly	Glu	Ile	Arg	Thr	Pro	Asp	Thr	
	145					150					155					160	
	CAA	TCT	GTG	CGA	ACT	ACA	TCA	GGT	CCT	TTG	GGT	CCT	TCT	GAC	AGG	AAT	528
	Gln	Ser	Val	Arg	Thr	Thr	Ser	Gly	Pro	Leu	Gly	Pro	Ser	Asp	Arg	Asn	
35					165					170					175		
			_														
												CCT					576
	Ala	Ile	Ser		Pro	Tyr	Ile	Asp		Arg	Gln	Pro	Val		Val	Arg	
				180					185					190			

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	ATC	GTG	GAC	CCG	TCA	AAA	GAC	TTG	AAC	TCT	TAT	GGG	CTT	GGT	AAT	GTT	624
	Ile	Val	Asp	Pro	Ser	Lys	Asp	Leu	Asn	Ser	Tyr	Gly	Leu	Gly	Asn	Val	
			195					200					205				
5	GAC	TGG	AAA	GAA	AGA	GTT	GAA	GGC	TGG	AAG	CTG	AAG	CAG	GAG	AAA	AAT	672
	Asp	Trp	Lys	Glu	Arg	Val	Glu	Gly	Trp	Lys	Leu	Lys	Gln	Glu	Lys	Asn	
		210					215					220					
	ATG	TTA	CAG	ATG	ACT	GGT	AAA	TAC	CAT	GAA	GGG	AAA	GGA	GGA	GAA	TTA	720
10	Met	Leu	Gln	Met	Thr	Gly	Lys	Tyr	His	Glu	Gly	Lys	Gly	Gly	Glu	Ile	
	225					230					235					240	
	GAA	GGG	ACT	GGT	TCC	AAT	GGC	GAA	GAA	CTC	CAA	ATG	GCT	GAT	GAT	ACA	768
	Glu	Gly	Thr	Gly	Ser	Asn	Gly	Glu	Glu	Leu	Gln	Met	Ala	Asp	Asp	Thr	
15					245					250					255		
	CGT	CTT	CCT	ATG	AGT	CGT	GTG	GTG	CCT	ATC	CCA	TCT	TCT	CGC	CTA	ACC	816
	Arg	Leu	Pro	Met	Ser	Arg	Val	Val	Pro	Ile	Pro	Ser	Ser	Arg	Leu	Thr	
				260					265					270			
20																	
	CCT	TAT	CGG	GTT	GTG	ATT	ATT	CTC	CGG	CTT	ATC	ATC	TTG	TGT	TTC	TTC	864
	Pro	Tyr	Arg	Val	Val	Ile	Ile	Leu	Arg	Leu	Ile	Ile	Leu	Cys	Phe	Phe	
			275					280					285				
25	TTG	CAA	TAT	CGT	ACA	ACT	CAC	CCT	GTG	AAA	AAT	GCA	TAT	CCT	TTG	TGG	912
	Leu	Gln	Tyr	Arg	Thr	Thr	His	Pro	Val	Lys	Asn	Ala	Tyr	Pro	Leu	Trp	
		290					295					300					
				GTT													960
30	Leu	Thr	Ser	Val	Ile	CAa	Glu	Ile	Trp	Phe	Ala	Phe	Ser	Trp	Leu	Leu	
	305					310					315					320	
	GAT	CAG	TTT	CCC	AAA	TGG	TAC	CCC	ATT	AAC	AGG	GAG	ACT	TAT	CTT	GAC	1008
	Yab	Gln	Phe	Pro	Lys	Trp	Tyr	Pro	Ile	Asn	Arg	Glu	Thr	Tyr	Leu	Asp	
35					325					330					335		
	CGT	CTC	GCT	ATA	AGA	TAT	GAT	CGA	GAC	GGT	GAA	CCA	TCA	CAG	CTC	GTT	1056
	Arg	Leu	Ala	Ile	Arg	Tyr	Asp	Arg	Asp	Gly	Glu	Pro	Ser	Gln	Leu	Val	
				340					345					350			
40																	

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	CCT	GTT	GAT	GTG	TTT	GTT	AGT	ACA	GTG	GAC	CCA	TTG	AAA	GAG	CCT	ccc	1104
	Pro	Val	Asp	Val	Phe	Val	Ser	Thr	Val	Asp	Pro	Leu	Lys	Glu	Pro	Pro	
			355					360					365				
5	CTT	GTT	ACA	GCA	AAC	ACA	GTT	CTC	TCG	ATT	CTT	TCT	GTG	GAC	TAC	CCG	1152
	Leu	Val	Thr	Ala	Asn	Thr	Val	Leu	Ser	Ile	Leu	Ser	Val	Asp	Tyr	Pro	
		370					375					380					
	GTA	GAT	AAA	GTA	GCC	TGT	TAT	GTT	TCA	GAT	GAT	GGT	TCA	GCT	ATG	CTT	1200
10	Val	Asp	Lys	Val	Ala	Cys	Tyr	Val	Ser	Asp	Asp	Gly	Ser	Ala	Met	Leu	
	385					390					395					400	
	ACC	TTT	GAA	TCC	CTT	TCT	GAA	ACC	GCT	GAG	TTT	GCA	AAG	AAA	TGG	GTA	1248
	Thr	Phe	Glu	Ser	Leu	Ser	Glu	Thr	Ala	Glu	Phe	Ala	Lys	Lys	Trp	Val	
15					405					410					415		
	CCA	TTT	TGC	AAG	AAA	TTC	AAC	ATT	GAA	CCT	AGG	GCC	CCT	GAA	TTC	TAT	1296
	Pro	Phe	Сув	Lys	Lys	Phe	Asn	Ile	Glu	Pro	Arg	Ala	Pro	Glu	Phe	Tyr	
••				420					425					430			
20																	
						GAT											1344
	Phe	Ala	Gln	Lys	Ile	Asp	Tyr	Leu	Lys	Asp	Lys	Ile	Gln	Pro	Ser	Phe	
			435					440					445				
25																	
25	GTT																1392
	Val		Glu	Arg	Arg	Ala		Lys	Arg	Glu	Tyr	Glu	Glu	Phe	Lys	Val	
		450					455					460					
20						GTT											1440
30	Arg	He	Asn	Ala	Leu		Ala	Lys	Ala	Gln		Ile	Pro	Glu	Glu	Gly	
	465					470					475					480	
	<b>500</b>																
						GGT											1488
35	Trp	Tnr	Met	GIN		Gly	Thr	Pro	Trp		Gly	Asn	Asn	Thr		Asp	
33					485					490					495		
	0.5	-															
						CAG											1536
	H18	PTO	GIY		116	Gln	val	Pne		GTÅ	His	Ser	Gly	-	Leu	Asp	
40				500					505					510			
40																	

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	ACC	GAT	GGA	AAT	GAG	CTG	CCT	AGA	CTC	ATC	TAT	GTT	TCT	CGT	GAA	AAG	1584
	Thr	Asp	Gly	Asn	Glu	Leu	Pro	Arg	Leu	Ile	Tyr	Val	Ser	Arg	Glu	Lys	
			515					520					525				
5	CGG	CCT	GGA	TTT	CAA	CAC	CAC	AAA	AAG	GCT	GGA	GCT	ATG	TAA	GCA	TTG	1632
	Arg	Pro	Gly	Phe	Gln	His	His	Lys	Lys	Ala	Gly	Ala	Met	Asn	Ala	Leu	
		530					535					540					
	ATC	CGT	GTA	TCT	GCT	GTT	CTT	ACC	AAT	GGA	GCA	TAT	CTT	TTG	AAC	GTG	1680
10	Ile	Arg	Val	Ser	Ala	Val	Leu	Thr	Asn	Gly	Ala	Tyr	Leu	Leu	Asn	Val	
	545					\$50					555					560	
	GAT	TGT	GAT	CAT	TAC	TTT	AAT	AAC	AGT	AAG	GCT	ATT	AAA	GAA	GCT	ATG	1728
	Asp	Сув	Asp	His	Tyr	Phe	Asn	Asn	Ser	Lys	Ala	Ile	Lys	Glu	Ala	Met	
15					565					570					<b>57</b> 5		
				ATG													1776
	Cys	Phe	Met	Met	qeA	Pro	Ala	Ile	Gly	Lys	Lys	Сув	Сув	Tyr	Val	Gln	
20				580					585					590			
20																	
				CGT													1824
	Phe	Pro		Arg	Phe	Asp	Gly		Ąsp	Leu	His	Asp	_	Tyr	Ala	Asn	
			595					600					605				
25																	
25				GTC													1872
	Arg		iie	Val	Phe	Phe	-	He	Asn	Met	Lys	-	Leu	Asp	GIY	Ile	
		610					615					620					
	CVC	COT	CCA	GTA	ጥለጥ	CTC	CCT	л Ст	COT	m~m	Tr.C.Tr	an an an	5 5 T	NCC	CNC	CCT	1920
30				Val													1920
-	625	Oly		vai	.y.	630	Gly	****	GIY	Cys	635	FILE	ASII	Arg	GIN	640	
						050					033					040	
	CTA	TAT	GGG	TAT	GAT	ССТ	GTT	ттс	ACG	GAA	GAA	GAT	מייינ	GAA	CCA	ДДТ	1968
				Tyr										•			1,00
35		•	-,		645					650					655		
	ATT	ATT	GTC	AAG	AGC	TGT	TGC	GGG	TCA	AGG	AAG	AAA	GGT	AAA	AGT	AGC	2016
				Lys													
				660		-	•	•	665	-	-	•	•	670			
40																	

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	AAG	AAG	TAT	AAC	TAC	GAA	AAG	AGG	AGA	GGC	ATC	AAC	AGA	AGT	GAC	TCC	2064
	Lys	Lys	Tyr	Asn	Tyr	Glu	Lys	Arg	Arg	Gly	Ile	Asn	Arg	Ser	Asp	Ser	
			675					680					685				
5	TAA	GCT	CCA	CTT	TTC	AAT	ATG	GAG	GAC	ATC	GAT	GAG	GGT	TTT	GAA	GGT	2112
	Asn	Ala	Pro	Leu	Phe	Asn	Met	Glu	Asp	Ile	Asp	Glu	Gly	Phe	Glu	Gly	
		690					695					700					
	TAT	GAT	GAT	GAG	AGG	TCT	ATT	CTA	ATG	TCC	CAG	AGG	AGT	GTA	GAG	AAG	2160
10	Tyr	Asp	Asp	Glu	Arg	Ser	Ile	Leu	Met	Ser	Gln	Arg	Ser	Val	Glu	Lys	
	705					710					715					720	
	CGT	TTT	GGT	CAG	TCG	CCG	GTA	TTT	ATT	GCG	GCA	ACC	TTC	ATG	GAA	CAA	2208
	Arg	Phe	Gly	Gln	Ser	Pro	Val	Phe	Ile	Ala	Ala	Thr	Phe	Met	Glu	Gln	
15					725					730					735		
	GGC	GGC	ATT	CCA	CCA	ACA	ACC	AAT	CCC	GCT	ACT	CTT	CTG	AAG	GAG	GCT	2256
	Gly	Gly	Ile	Pro	Pro	Thr	Thr	Asn	Pro	Ala	Thr	Leu	Leu	Lys	Glu	Ala	
				740					745					750			
20																	
	TTA	CAT	GTT	ATA	AGC	TGT	GGT	TAC	GAA	GAC	AAG	ACT	GAA	TGG	GGC	AAA	2304
	Ile	His	Val	Ile	Ser	Cys	Gly	Tyr	Glu	Asp	Lys	Thr	Glu	Trp	Gly	Lys	
			755					760					765				
25				TGG													2352
	Glu	Ile	Gly	Trp	Ile	Tyr	Gly	Ser	Val	Thr	Glu	Asp	Ile	Leu	Thr	Gly	
		770					775					780					
••				CAT													2400
30	Phe	Lys	Met	His	Ala	Arg	Gly	Trp	Ile	Ser	Ile	Tyr	Сув	Asn	Pro	Pro	
	785					790					795					800	
				TTC													2448
2.0	Arg	Pro	Ala	Phe	ГÅЗ	Gly	Ser	Ala	Pro	Ile	Asn	Leu	Ser	qaA	Arg	Leu	
35					805					810					815		
				CTT													2496
	Asn	Gln	Val	Leu	Arg	Trp	Ala	Leu	_	Ser	Ile	Glu	Ile		Leu	Ser	
				820					825					830			

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	AGA	CAT	TGT	CCT	ATC	TGG	TAT	GGT	TAC	CAT	GGA	AGG	TTG	AGA	CTT	TTG	2544
	Arg	His	Cys	Pro	Ile	Trp	Tyr	Gly	Tyr	His	Gly	Arg	Leu	Arg	Leu	Leu	
			835					840					845				
5	GAG	AGG	ATC	GCT	TAT	ATC	AAC	ACC	ATC	GTC	TAT	CCT	TTA	ACA	TCC	ATC	2592
	Glu	Arg	Ile	Ala	Tyr	Ile	Asn	Thr	Ile	Val	Tyr	Pro	Ile	Thr	Ser	Ile	
		850					855					860					
	CCT	CTT	ATT	GCG	TAT	TGT	ATT	CTT	CCC	GCT	TTT	TGT	CTC	ATC	ACC	GAC	2640
0	Pro	Leu	Ile	Ala	Tyr	Cys	Ile	Leu	Pro	Ala	Phe	Сув	Leu	Ile	Thr	Asp	
	865					870					B75					880	
	AGA	TTC	ATC	ATA	CCC	GAG	ATA	AGC	AAC	TAC	GCG	AGT	ATT	TGG	TTC	ATT	2688
	Arg	Phe	lle	Ile	Pro	Glu	Ile	Ser	neA	Tyr	Ala	Ser	Ile	Trp	Phe	Ile	
15					885					890					895		
	CTA	CTC	TTC	ATC	TCA	ATT	GCT	GTG	ACT	GGA	ATC	CTG	GAG	CTG	AGA	TGG	2736
	Leu	Leu	Phe	Ile	Ser	Ile	Ala	Val	Thr	Gly	Ile	Leu	Glu	Leu	Arg	Trp	
				900					905					910			
20																	
	AGC	GGT	GTG	AGC	ATT	GAG	GAT	TGG	TGG	AGG	AAC	GAG	CAG	TTC	TGG	GTC	2784
	Ser	Gly	Val	Ser	Ile	Glu	Asp	Trp	Trp	Arg	Asn	Glu	Gln	Phe	Trp	Val	
			915					920					925				
25	ATT	GGT	GGC	ACA	TCC	GCC	CAT	CTT	TTT	GCT	GTC	TTC	CAA	GGT	CTA	CTT	2832
	Ile	Gly	Gly	Thr	Ser	Ala	His	Leu	Phe	Ala	Val	Phe	Gln	Gly	Leu	Leu	
		930					935					940					
										-	ACC			-			2880
30	Lys	Val	Leu	Ala	Gly	Ile	Asp	Thr	Asn	Phe	Thr	Val	Thr	Ser	Lys	Ala	
	945					950					955					960	
	ACA	GAC	GAA	GAT	GGG	GAT	TTT	GCA	GAA	CTC	TAC	ATC	TTC	AAA	TGG	ACA	2928
	Thr	Asp	Glu	Asp	Gly	Asp	Phe	Ala	Glu	Leu	Tyr	Ile	Phe	Lys	Trp	Thr	
35					965					970					975		
											CTT						2976
	Ala	Leu	Leu	Ile	Pro	Pro	Thr	Thr	Val	Leu	Leu	Val	Asn	Leu	Ile	Gly	
				980					985					990			

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	TTA	GTG	GCT	GGT	GTC	TCT	TAT	GCT	GTA	AAC	AGT	GGC	TAC	CAG	TCG	TGG	3024
	Ile	Val	Ala	Gly	Val	Ser	Tyr	Ala	Val	Asn	Ser	Gly	Tyr	Gln	Ser	Trp	
			995					1000	)				1009	5			
5	GGT	CCG	CTT	TTC	GGG	AAG	CTC	TTC	TTC	GCC	ATT	TGG	GTT	ATT	GCC	CAT	3072
	Gly	Pro	Leu	Phe	Gly	Lys	Leu	Phe	Phe	Ala	Leu	Trp	Val	Ile	Ala	His	
		1010	)				1019	5				1020	)				
	CTC	TAC	CCT	TTC	TTG	AAA	GGT	CTG	TTG	GGA	AGA	CAA	AAC	CGA	ACA	CCA	3120
10	Leu	Tyr	Pro	Phe	Leu	Lys	Gly	Leu	Leu	Gly	Arg	Gln	Asn	Arg	Thr	Pro	
	1025	5				1030	)				103	5				1040	
	ACC	ATC	GTC	TTA	GTC	TGG	TCT	GTT	СТТ	CTC	GCC	TCC	ATC	TTC	TCG	TTG	3168
	Thr	Ile	Val	Ile	Val	Trp	Ser	Val	Leu	Leu	Ala	Ser	Ile	Phe	Ser	Leu	
15					1049					1050					105		
	CTT	TGG	GTC	AGG	ATC	AAT	ccc	TTT	GTG	GAC	GCC	AAT	ccc	AAT	GCC	AAC	3216
	Leu	Trp	Val	Arg	Ile	Asn	Pro	Phe	Val	Asp	Ala	Asn	Pro	Asn	Ala	Asn	
		•		1060					1069	•				1076			
20																	
	AAC	TTC	AAT	GGC	AAA	GGA	GGT	GTC	TTT	TAG	ACCC	TAT 1	ГТАТ	ATAC	гт		3263
				Gly													5500
			107	•	-7-	,	,	1080									
				•					•								
25	GTGT	ויניזיני	ግልጥ :	ነጥ ልጥ ል	מממ~	<b>22 C</b> (	arar:	ስ አጥርረ	י מיט	ስ ተጥር የ	מממ~	ידריאי	ኮሮሞል:	ממ	~CAT	CAAACC	3323
	0.0.	.010					3000	<b>1110</b> 0	, 0,72	1110		100	i C i Au	<b></b>			3323
	CCN	י מיישיר	NCC (	ייייי	እ <i>ር</i> ጥጥ:	N	יתיי אי	ויייררי	, mor	rccn:		TRO	ammer (	ome (	~~ × ~	TAGCCA	3383
	CCA	JIGM	ACC (	3666	4.6111	MA G	JIGA	1100	4 IG.	ICCA	HGMI	IAG	C111\	-10	LUAU	IAGCCA	3363
	CNC	אממי	TC A	N N ጥጥ/	~ TTTC	ግጥ እ		T R (1414	י מידו	n m~ n	naan	CON	7maa	20 h		GATGTG	3443
30	GAG	-UNGG	IGA A	MAI I	3110	JI A	HCHC.	IMII	3 1A/	HIGA	1111	CCA	3166	JGA A	HUAA	BAIGIG	3443
50	03.00	7(13 B :	N TO C		. m	ma m											2502
	GAC	CAA	AIG A	ATAC	ATAG	re T	ACAA	AAAG/	A AT	l'IGT	IAI I	CTT	rett	ATA '	I'I'IA	TTTTAT	3503
	TTA	HAGC"	TIG '	TAG	ACTC	AC A	CLLY.	IGTA	A TG	r rGG	AACT	TGT'	IGTC	CTA I	AAAA	GGGATT	3563
25								nas - :									
دد	GGA(	"I"I"!	rct '	TTTT	ATCT	AA G	AATC	rgaa(	; TT	ľATA'	IGCT						3603

(2) INFORMATION FOR SEQ ID NO:6:

40 (i) SEQUENCE CHARACTERISTICS:

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(A) LENGTH: 1081 amino acids

				(B)	TY	PE: a	amino	ac:	id							
				(D)	TO	POLO	3Y: :	linea	ar							
5		(:	ii) l	MOLE	CULE	TYPI	E: p	rote:	in							
		()	ki} S	SEQUI	ENCE	DES	CRIP	rion	: SE(	Q ID	NO:	5 :				
10	Met 1	Glu	Ala	Ser	Ala 5	Gly	Leu	Val	Ala	Gly 10	Ser	Tyr	Arg	Arg	Asn 15	Glu
	Leu	Val	Arg	Ile 20	Arg	Н18	Glu	Ser	Asp 25	Gly	Gly	Thr	Lys	Pro 30	Leu	Lys
15	Asn	Met	Asn 35	Gly	Gln	Ile	Суз	Gln 40	Ile	Сув	Gly	Ąsp	Asp 45	Val	Gly	Leu
20	Ala	Glu 50	Thr	Gly	Asp	Val	Phe 55	Val	Ala	Сув	Asn	Glu 60	Сув	Ala	Phe	Pro
	Val 65	Cys	Arg	Pro	Cys	Tyr 70	Glu	Tyr	Glu	Arg	Lys 75	Asp	Gly	Thr	Gln	Cys 80
25	Cys	Pro	Gln	аүЭ	Lys 85	Thr	Arg	Phe	Arg	Arg 90	His	Arg	Gly	Ser	Pro 95	Arg
	Val	Glu	Gly	Asp	Glu	Asp	Glu	Asp	Asp 105	Val	Asp	Asp	Ile	Glu 110	Asn	Glu
30	Phe	Asn	Tyr 115	Ala	Gln	Gly	Ala	Asn 120	Lys	Ala	Arg	His	Gln 125	Arg	His	Gly
35	Glu	Glu 130	Phe	Ser	Ser	Ser	Ser 135	Arg	His	Glu	Ser	Gln 140	Pro	Ile	Pro	Leu
	Leu 145	Thr	His	Gly	His	Thr 150	Val	Ser	Gly	Glu	Ile 155	Arg	Thr	Pro	qaA	Thr 160
40	Gln	Ser	Val	Arg	Thr 165	Thr	Ser	Gly	Pro	Leu 170	Gly	Pro	Ser	Asp	Arg 175	Asn

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	Ala	Ile	Ser	Ser	Pro	Tyr	Ile	Asp	Pro	Arg	Gln	Pro	Val	Pro	Val	Arg
				180					185					190		
	Ile	Val	Asp	Pro	Ser	Lys	Asp	Leu	Asn	Ser	Tyr	Gly	Leu	Gly	Asn	Val
5			195					200					205			
	Asp	Trp	Lys	Glu	Arg	Val	Glu	Gly	Trp	Lys	Leu	Lys	Gln	Glu	Lys	Asn
		210					215					220				
10	Met	Leu	Gln	Met	Thr	Gly	Lys	Tyr	His	Glu	Gly	Lys	Gly	Gly	Glu	Ile
	225					230					235					240
	Glu	Gly	Thr	Gly	Ser	Asn	Gly	Glu	Glu	Leu	Gln	Met	Ala	Asp	Asp	Thr
					245					250					255	
15																
	Arg	Leu	Pro	Met	Ser	Arg	Val	Val	Pro	Ile	Pro	Ser	Ser	Arg	Leu	Thr
				260					265					270		
	Pro	Tyr	Arg	Val	Val	Ile	Ile	Leu	Arg	Leu	Ile	Ile	Leu	Cys	Phe	Phe
20			275					280					285			
	Leu	Gln	Tyr	Arg	Thr	Thr	His	Pro	Val	Lys	Asn	Ala	Tyr	Pro	Leu	Trp
		290					295					300				
25	Leu	Thr	Ser	Val	Ile	Сув	Glu	Ile	Trp	Phe	Ala	Phe	Ser	Trp	Leu	Leu
	305					310					315			-		320
	Asp	Gln	Phe	Pro	Lys	Trp	Tyr	Pro	Ile	Asn	Arg	Glu	Thr	Tyr	Leu	Asp
					325					330					335	
30																
	Arg	Leu	Ala	Ile	Arg	Tyr	Asp	Arg	Asp	Gly	Glu	Pro	Ser	Gln	Leu	Val
				340					345					350		
	Pro	Val	Asp	Val	Phe	Val	Ser	Thr	Val	Авр	Pro	Leu	Lys	Glu	Pro	Pro
35			355					360		_			365			
	Leu	Val	Thr	Ala	Asn	Thr	Val	Leu	Ser	Ile	Leu	Ser	Val	αaA	Tyr	Pro
		370					375					380			- 4 -	

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	Val 385	Asp	Lys	Val	Ala	390	Tyr	Val	Ser	Asp	395	Gly	Ser	Ala	Met	<b>L</b> ev 400
4		Phe	Glu	Ser	Leu 405	Ser	Glu	Thr	Ala	Glu 410	Phe	Ala	Lys	Lys	Trp 415	Val
	Pro	Phe	Cys	Lys 420	Lys	Phe	Asn	Ile	Glu 425	Pro	Arg	Ala	Pro	Glu 430	Phe	Туг
10	) Phe	Ala	Gln 435	Lys	Ile	Asp	Tyr	Leu 440	Lys	Asp	Lys	Ile	Gln 445	Pro	Ser	Phe
15		Lys 450	Glu	Arg	Arg	Ala	Met 455	Lys	Arg	Glu	Tyr	Glu 460	Glu	Phe	Lys	Val
		Ile	Asn	Ala	Leu	Val 470	Ala	Lys	Ala	Gln	<b>L</b> ув 475	Ile	Pro	Glu	Glu	Gly 480
20	_	Thr	Met	Gln	Asp 485	Gly	Thr	Pro	Trp	Pro 490	Gly	Asn	Asn	Thr	Arg 495	Asp
	His	Pro	Gly	Met 500	Ile	Gln	Val	Phe	Leu 505	Gly	His	Ser	Gly	Gly 510	Leu	Asp
25	Thr	Asp	Gly 515	Asn	Glu	Leu	Pro	Arg 520	Leu	Ile	туг	Val	Ser 525	Arg	Glu	Lys
30		Pro 530	Gly	Phe	Gln	His	His 535	Lys	Lys	Ala	Gly	Ala 540	Met	Asn	Ala	Leu
	Ile 545	Arg	Val	Ser	Ala	Val 550	Leu	Thr	Asn	Gly	Ala 555	Tyr	Leu	Leu	Asn	Val
35		Cys	Asp	His	Tyr 565	Phe	Asn	Asn	Ser	Lys 570	Ala	Ile	Lys	Glu	Ala 575	Met
	Сув	Phe	Met	Met 580	Авр	Pro	Ala	Ile	Gly 585	Lys	Lys	Сув	Сув	Tyr 590	Val	Gln

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	Pne	PIO	595	Arg	Pne	Asp	GIY	600	Asp	rea	uis	Asp	605	Tyr	Ala	ASN
	Arg	Asn	Ile	Val	Phe	Phe	Asp	Ile	Asn	Met	Lys	Gly	Leu	Asp	Gly	Ile
5		610					615					620				
	Gln	Gly	Pro	Val	Tyr	Val	Gly	Thr	Gly	Cys	Cys	Phe	Asn	Arg	Gln	
	625					630					635					640
10	Leu	Tyr	Gly	Tyr	Asp 645	Pro	Val	Leu	Thr	Glu 650	Glu	Asp	Leu	Glu	Pro	Asn
						_	_									_
	Ile	Ile	Val	660 Lys	Ser	Сув	Сув	Gly	Ser 665	Arg	Lys	Lys	Gly	Lys 670	Ser	Ser
15	Lys	Lys	Tyr	Asn	Tyr	Glu	Lys	Arg	Arg	Gly	Ile	Asn	Arg	Ser	Asp	Ser
	•	•	675		-		-	680	_				685		•	
	Asn	Ala	Pro	Leu	Phe	Asn	Met	Glu	Asp	Ile	Asp	Glu	Gly	Phe	Glu	Gly
20		690					695					700				
	Tyr 705	Asp	Asp	Glu	Arg	Ser 710	Ile	Leu	Met	Ser	Gln 715	Arg	Ser	Val	Glu	Lys 720
25															0.2	
23	Arg	Phe	Gly	Gln	Ser 725	Pro	Val	Phe	Ile	730	Ala	Thr	Phe	Met	Glu 735	Gln
	Gly	Gly	Ile	Pro	Pro	Thr	Thr	Asn	Pro	Ala	Thr	Leu	Leu	Lys	Glu	Ala
30				740					745					750		
50	Ile	His		Ile	Ser	Cys	Gly			Asp	Lys	Thr		Trp	Gly	Lys
			755					760					765			
35		Ile 770	_	Trp	Ile	Tyr	Gly 775	Ser	Val	Thr	Glu	Asp 780	Ile	Leu	Thr	Gly
		1	Mak	n + ~	<b>N</b> 3-	A	G3	Τ⊶	T1-	Ca		T	C	N	D=-	D=-
	785	-	ייפנ	His	WIG	790	_	rrp	116	JeI	795	_	Cys	URII	FLO	800

	Arg	Pro	Ala	Phe	-	Gly	Ser	Ala	Pro		Asn	Leu	Ser	Asp	-	Let
					805					810					815	
	Asn	Gln	Val	Leu	Arg	Trp	Ala	Leu	Gly	Ser	Ile	Glu	Ile	Leu	Leu	Ser
5				820					825					830		
	Arg	His	Cys	Pro	Ile	Trp	Tyr	Gly	Tyr	His	Gly	Arg	Leu	Arg	Leu	Leu
			835					840	Ū			_	845			
10	-1				_			_,			_	_			_	
10	Glu	850	Ile	Ala	Tyr	He	Asn 855	Thr	He	vai	туг	860	He	Thr	Ser	Ile
		Leu	Ile	Ala	Tyr		Ile	Leu	Pro	Ala		Cys	Leu	Ile	Thr	
15	865					870					875					880
	Arg	Phe	Ile	Ile	Pro	Glu	Ile	Ser	Asn	Tyr	Ala	Ser	Ile	Trp	Phe	Ιlε
					885					890					895	
	Leu	Leu	Phe	Ile	Ser	Ile	Ala	Val	Thr	Gly	Ile	Leu	Glu	Leu	Arg	Trp
20				900					905					910		
	Co	G)	v.1	Co	T1.	C1	N	T.	Т	2	N ===	<b>01</b>	G) =	nh.	<b>6</b> 0	V-1
	ser	GIY	Val 915	ser	11e	GIU	Asp	920	Trp	Arg	Asn	GIU	925	Pne	Trp	vai
25	Ile	_	Gly	Thr	Ser	Ala		Leu	Phe	Ala	Val		Gln	Gly	Leu	Leu
		930					935					940				
	Lys	Val	Leu	Ala	Gly	Ile	Asp	Thr	Asn	Phe	Thr	Val	Thr	Ser	Lys	Ala
30	945					950					955					960
50	Thr	Asp	Glu	Asp	Gly	Asp	Phe	Ala	Glu	Leu	Tyr	Ile	Phe	Lys	Trp	Thr
					965					970				-	975	
	<b>71</b> ~	Lev	Levi	110	Dro	Dro	ሞኮ~	Th~	Vo 1	T.c.	T.c.	U-1	<b>N</b> ==	T a	<b>+1</b> -	<b>C</b> 1-
35	nra.	neu	Leu	980	F10	FIO	THE	1111	985	neu	Deu	val	ASII	990	116	GIÀ
	Ile	Val	Ala	Gly	Val	Ser	Tyr			Asn	Ser				Ser	Trp
			995					1000	ı				1009	•		

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Gly Pro Leu Phe Gly Lys Leu Phe Phe Ala Leu Trp Val Ile Ala His 1010 1015 1020

Leu Tyr Pro Phe Leu Lys Gly Leu Leu Gly Arg Gln Asn Arg Thr Pro 5 1025 1030 1035 1040

Thr Ile Val Ile Val Trp Ser Val Leu Leu Ala Ser Ile Phe Ser Leu 1045 1050 1055

10 Leu Trp Val Arg Ile Asn Pro Phe Val Asp Ala Asn Pro Asn Ala Asn 1060 1065 1070

Asn Phe Asn Gly Lys Gly Gly Val Phe 1075 1080

15

- (2) INFORMATION FOR SEQ ID NO:7:
- 20 (i) SEQUENCE CHARACTERISTICS:

(A) LENGTH: 3828 base pairs

(B) TYPE: nucleic acid

(C) STRANDEDNESS: single

(D) TOPOLOGY: linear

25

- (ii) MOLECULE TYPE: cDNA
- (iii) HYPOTHETICAL: NO
- 30 (iv) ANTI-SENSE: NO
  - (vi) ORIGINAL SOURCE:
    - (A) ORGANISM: Arabidopsis thaliana
    - (B) STRAIN: Columbia

- (vii) IMMEDIATE SOURCE:
  - (B) CLONE: Ath-A
- (ix) FEATURE:
- 40 (A) NAME/KEY: CDS

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## (B) LOCATION: 239..3490

		(xi)	SEC	QUENC	CE DE	ESCR	IPTIC	ON: S	SEQ :	ID NO	):7:						
5	GTC	GACAC	CTA A	AGTGC	SATCO	EA AZ	AGAA:	TCG	C GG	cccc	STCG	ATA	CGC	rgc (	GAGA	AGACGA	60
	CAG	<b>AA</b> GG(	GGA 1	rtgto	CGAT	rc go	3TTT	ATTT(	C GT	CTCC	rtcg	TCT	rcca(	CTC :	rtac"	<b>FAGTGC</b>	120
10	ATG	CTCTC	GAA 1	rctgi	ratg:	ΓΑ ΑΊ	rgggi	AGTT(	C AA	CAGT	CTGG	ATC	CATT	ATC (	CTAG	CCGGGT	180
	CGGC	STCAJ	AGG 1	rctt	rgaa7	ra ac	BAGA	GACAI	A TT	CGTT	rtga	TTC	GTGT	rag i	<b>A</b> AGA(	CATC	238
				GGT													286
15	Met	Asn	Thr	Gly	Gly	Arg	Leu	Ile	Ala	Gly	Ser	His	Asn	Arg	Asn	Glu	
	1				5					10					15		
	TTC	GTT	CTC	ATT	AAC	GCC	GAT	GAG	AGT	GCC	AGA	ATA	CGA	TCA	GTA	CAA	334
••	Phe	Val	Leu	Ile	Asn	Ala	Asp	Glu	Ser	Ala	Arg	Ile	Arg	Ser	Val	Gln	
20				20					25					30			
				GGG													382
	Glu	Leu		Gly	Gln	Thr	Cys		Ile	Сув	Gly	Asp		Ile	Glu	Leu	
25			35					40					45				
23		-															
				AGT													430
	inr	50	ser	Ser	GIU	Leu		vai	Ala	Cys	Asn		Cys	AIS	Pne	Pro	
		50					55					60					
30	CTT	тст	AGA	CCA	TGC	ጥልጥ	GAG	יימיי	GAA	ССТ	DCD.	CAA	CCA	ידממ	CAA	CCT	478
				Pro													• • • • • • • • • • • • • • • • • • • •
	65	0,0			-,-	70	014	-,-	0.0	9	75	014	O <sub>1</sub>	Au	01	80	
	TGT	CCT	CAG	TGC	AAA	ACT	CGA	TAC	AAA	AGG	ATT	AAA	GGT	AGT	CCA	CGG	526
35	Сув	Pro		Сув													
	-			•	85		-	•		90		•	-		95	•	
	GTT	GAT	GGA	GAT	GAT	GAA	GAA	GAA	GAA	GAC	ATT	GAT	GAT	CTT	GAG	TAT	574
	Val	Asp	Gly	Asp	Asp	Glu	Glu	Glu	Glu	Asp	Ile	Asp	Asp	Leu	Glu	Tyr	
40				100					105					110			

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	GAG	TTT	GAT	CAT	GGG	ATG	GAC	ССТ	GAA	CAT	GCC	GCT	GAA	GCC	GCA	CTC	622
	Glu	Phe	qeA	His	Gly	Met	Asp	Pro	Glu	His	Ala	Ala	Glu	Ala	Ala	Leu	
			115					120					125				
5	TCT	TCA	CGC	CTT	AAC	ACC	GGT	CGT	GGT	GGA	TTG	GAT	TCA	GCT	CCA	CCT	670
	Ser	Ser	Arg	Leu	Asn	Thr	Gly	Arg	Gly	Gly	Leu	Asp	Ser	Ala	Pro	Pro	
		130					135					140					
	GGC	TCT	CAG	TTA	CCT	CTT	TTG	ACT	TAT	TGT	GAT	GAA	GAT	GCT	GAT	ATG	718
10	Gly	Ser	Gln	Ile	Pro	Leu	Leu	Thr	Tyr	Cys	Asp	Glu	Asp	Ala	Asp	Met	
	145					150					155					160	
															TAT		766
15	Tyr	Ser	Asp	Arg		Ala	Leu	Ile	Val		Pro	Ser	Thr	GIA	Tyr	Gly	
15					165					170					175		
		000	ama	m » m		000	200	mmm	n.c.n	~~m	more.	mom.			CC3	CNC	014
										-					CCA		814
	Asn	Arg	vaı	-	PIO	Ala	Pro	Pne		Asp	ser	ser	ATS		Pro	GIN	
20				180					185					190			
20	ccc	ממ	TCA	בעתה	COTO	CCT	CNG	***	ርአጥ	እጥጥ	ccc	ממט	ጥለጥ	COT	TAT	CCA	862
															Tyr		802
	710	w. A	195	MEC	Val	FIO	GIN	200	vob	116	NIG	GIU	205	GIY	ıyı	GLY	
			1,,					200					203				
25	AGT	GTT	GCT	TGG	AAG	GAC	CGT	ATG	GAA	GTT	TGG	AAG	AGA	CGA	CAA	GGC	910
															Gln		
		210		•		•	215					220				•	
	GAA	AAG	CTT	CAA	GTC	ATT	AAG	CAT	GAA	GGA	GGA	AAC	AAT	GGT	CGA	GGT	958
30	Glu	Lys	Leu	Gln	Val	Ile	Lys	His	Glu	Gly	Gly	Asn	Asn	Gly	Arg	Gly	
	225					230					235					240	
	TCC	AAT	GAT	GAC	GAC	GAA	CTA	GAT	GAT	CCT	GAC	ATG	CCT	ATG	ATG	GAT	1006
	Ser	Asn	Asp	Asp	Asp	Glu	Leu	Asp	Asp	Pro	Asp	Met	Pro	Met	Met	Asp	
35					245					250					255		
	GAA	GGA	AGA	CAA	CCT	CTC	TCA	AGA	AAG	CTA	CCT	ATT	CGT	TCA	AGC	AGA	1054
	Glu	Gly	Arg	Gln	Pro	Leu	Ser	Arg	Lys	Leu	Pro	Ile	Arg	Ser	Ser	Arg	
				260					265					270			

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	ATA	TAA	CCT	TAC	AGG	ATG	TTA	ATT	CTG	TGT	CGC	CTC	GCG	ATT	CTT	GGT	1102
	Ile	Asn	Pro	Tyr	Arg	Met	Leu	Ile	Leu	Cys	Arg	Leu	Ala	Ile	Leu	Gly	
			275					280					285				
5	CTT	TTC	TTT	CAT	TAT	AGA	ATT	CTC	CAT	CCA	GTC	AAT	GAT	GCA	TAT	GGA	1150
	Leu	Phe	Phe	His	Tyr	Arg	Ile	Leu	His	Pro	Val	Asn	Asp	Ala	Tyr	Gly	
		290					295					300					
	TTA	TGG	TTA	ACG	TCA	GTT	ATA	TGC	GAA	ATA	TGG	TTT	GCA	GTG	TCT	TGG	1198
10	Leu	Trp	Leu	Thr	Ser	Val	Ile	Суз	Glu	Ile	Trp	Phe	Ala	Val	Ser	Trp	
	305					310					315					320	
	ATT	CTT	GAT	CAA	TTC	CCC	AAA	TGG	TAT	CCT	ATA	GAA	CGT	GAA	ACA	TAC	1246
					Phe												
15			•		325		•	•	-	330					335	_	
	CTC	GAT	AGA	CTC	TCT	CTC	AGG	TAC	GAG	AAG	GAA	GGA	AAA	CCG	TCA	GGA	1294
					Ser												
				340			_	•	345	•		•	•	350		-	
20																	
	тта	GCA	ССТ	GTT	GAT	GTT	TTT	GTT	AGT	ACA	GTG	GAT	CCG	TTG	AAA	GAG	1342
					Asp												
			355					360					365		-2 -		
			•••														
25	רככ	רכר	ፐፐር	АТТ	ACA	GCA	AAC	ACA	GTT	CTT	TCC	АТТ	CTA	GCA	GTT	GAT	1390
					Thr												
	710	370	Deu	110	****	,,,,,	375		<b>741</b>	200	-	380	200		,,,	1.56	
		370					3,3					300					
	тдт	ררייי	GTG	GAT	AAG	<b>G</b> ጥጥ	GCG	<b>ፐር</b> ጥ	ТАТ	GTA	ፐርል	AAC	דממ	ርርጥ	GCA	GСT	1438
30					Lys												1130
50	385	110	VUI	nop	Lys	390		C) U	-1-	•	395	,,,,,,,	7.511	01,	*****	400	
	303					320					323					400	
	ስጥር	ىلىش	እሮአ	ידיידיי	GAA	COT	CTC	ጥርጣ	CAT	a Ca	CCT	CAT	Total	CCT	እሮአ	ממה	1486
					Glu												1400
35	MEL	Deu	1111	PILE	405	AIG	Deu	361	nsp	410	AIG	vah	FIIE	VIO	415	Буб	
,,					405					410					413		
	TCC.	രത്ത	COM	the treat	TGT	አአር	ልአሮ	ውውጥ	አአጥ	ልጥጣ	GAC	CCA	CCA	G/m	CCT	GAG	1534
																	1734
	rth	vai	PIO		Cys	nya	nya	FIIG	425	116	GIU	FIO	wrd		FIO	JIU	
40				420					423					430			
TU																	

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	TGG	TAT	TTT	TCT	CAG	AAG	ATG	GAT	TAC	CTG	AAG	AAC	AAA	GTT	CAT	CCT	1582
	Trp	Tyr	Phe	Ser	Gln	Lys	Met	Asp	Tyr	Leu	Lys	Asn	Lys	Val	His	Pro	
			435					440					445				
_																	
)												GAT					1630
	Ala		Val	Arg	Glu	Arg	_	Ala	Met	Lys	Arg	Asp	Tyr	Glu	Glu	Phe	
		450					455					460					
	מממ	GTG	אע	<b>አ</b> ሞአ	አአጥ	GCN	CTC	ርጥጥ	CCT	a cor	CCN	CAG	חחת	CTC.	CCT	CAC	1678
10												Gln					1070
	465		-,-			470					475		~,~			480	
	GAA	CGT	TGG	ACT	ATG	CAA	GAT	GGA	ACT	CCT	TGG	CCT	GGA	AAC	AAC	GTC	1726
	Glu	Arg	Trp	Thr	Met	Gln	Asp	Gly	Thr	Pro	Trp	Pro	Gly	Asn	Asn	Val	
15					485					490					495		
	CGT	GAC	CAT	CCT	GGA	ATG	ATT	CAG	GTG	TTC	TTG	GGT	CAT	AGT	GGA	GTT	1774
	Arg	Asp	His	Pro	Gly	Met	Ile	Gln	Val	Phe	Leu	Gly	His	Ser	Gly	Val	
• •				500					505					510			
20																	
												GTG					1822
	Arg	Asp		Asp	Gly	Asn	Glu		Pro	Arg	Leu	Val	•	Val	Ser	Arg	
			515					520					525				
25	CAC	220	ccc	COT	CCN	do do do	CAT	CAC	CAC	220	222	GCT	CCA	COT	አጥር፣	887	1870
23												Ala					1070
	GIU	530	Arg	710	Gry	FIIC	535	1110	1110	Dyo	Dys	540	Oly	A10	Mec	no	
												•					
	TCC	TTG	ATC	CGA	GTC	TCT	GCT	GTT	СТА	TCA	AAC	GCT	CCT	TAC	CTT	CTT	1918
30	Ser	Leu	Ile	Arg	Val	Ser	Ala	Val	Leu	Ser	Asn	Ala	Pro	Tyr	Leu	Leu	
	545					550					555					560	
	AAT	GTC	GAT	TGT	GAT	CAC	TAC	ATC	AAC	AAC	AGC	AAA	GCA	ATT	AGA	GAA	1966
	Asn	Val	Asp	Сув	Asp	His	Tyr	Ile	Asn	Asn	Ser	Lys	Ala	Ile	Arg	Glu	
35					565					570					575		
	TCT	ATG	TGT	TTC	ATG	ATG	GAC	CCG	CAA	TCG	GGA	AAG	AAA	GTT	TGT	TAT	2014
	Ser	Met	Сув	Phe	Met	Met	qaA	Pro	Gln	Ser	Gly	Lys	Lys	Val	Сув	Tyr	
				580					585					590			

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	GTT	CAG	TTT	CCG	CAG	AGA	TTT	GAT	GGG	ATT	GAT	AGA	CAT	GAT	AGA	TAC	2062
	Val	Gln	Phe	Pro	Gln	Arg	Phe	Asp	Gly	Ile	Asp	Arg	His	Asp	Arg	Tyr	
			595					600					605				
5	TCA	AAC	CGT	AAC	GTT	GTG	TTC	TTT	GAT	TTA	AAC	ATG	AAA	GGT	CTT	GAT	2110
	Ser	Asn	Arg	naA	Val	Val	Phe	Phe	yab	Ile	Asn	Met	Lys	Gly	Leu	Asp	
		610					615					620					
10												TGT					2158
10	_	Ile	Gin	GIY	Pro		Tyr	vaı	GIY	Thr		Сув	Val	Phe	Arg		
	625					630					635					640	
	CAG	CCT	CTT	тат	ССТ	ጥጥጥ	СУТ	GCA	CCA	AAG	AAG	AAG	444	CCA	CCA	GGC	2206
												Lys					2200
15	<b>02</b>		200	-,-	645					650	<b>-</b> 17-	2,0	_,_		655	,	
	AAA	ACC	TGT	AAC	TGT	TGG	CCT	AAA	TGG	TGT	TGT	TTG	TGT	TGT	GGG	TTG	2254
	Lув	Thr	Сув	Asn	Cys	Trp	Pro	Lys	Trp	Сув	Сув	Leu	Сув	Суз	Gly	Leu	
	•			660	-	•		-	665	•	-		•	670	•		
20																	
	AGA	AAG	AAG	AGT	AAA	ACG	AAA	GCC	ACA	GAT	AAG	AAA	ACT	AAC	ACT	AAA	2302
	Arg	Lys	Lys	Ser	Lys	Thr	Lys	Ala	Thr	Asp	Lys	Lys	Thr	Asn	Thr	Lys	
			675					680					685				
25	GAG	ACT	TCA	AAG	CAG	ATT	CAT	GCG	CTA	GAG	AAT	GTC	GAC	GAA	GGT	GTT	2350
	Glu	Thr	Ser	Lys	Gln	Ile	His	Ala	Leu	Glu	Asn	Val	Asp	Glu	Gly	Val	
		690					695					700					
20												GAA					2398
30		Val	Pro	Val	Ser		Val	Glu	Lys	Arg	Ser	Glu	Ala	Thr	Gln	Leu	
	705					710					715					720	
		-															
												TTC					2446
35	ràe	Leu	GIU	Lys	_	Pne	GIY	GIN	ser		vaı	Phe	vai	Ala		Ala	
33					725					730					735		
	CTT	СТР	CAG	244	GGT	CCP	ىلىلىت	כככ	CCT	ממ	GC»	AGC	CCC	GCP	יויבאנ	<b>ጥ</b> ር2	2494
												Ser					6777
				740	7	,			745		*****			750	Cyo	204	
40																	

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	TTA	AGA	GAA	GCC	ATT	CAA	GTT	ATT	AGC	TGC	GGG	TAC	CAA	GAT	AAA	ACC	2542
	Leu	Arg	Glu	Ala	Ile	Gln	Val	Ile	Ser	Cys	Gly	Tyr	Gln	Asp	Lys	Thr	
			755					760					765				
5	GAA	TGG	GGA	AAA	GAG	ATC	GGG	TGG	ATT	TAT	GGA	TÇG	GTG	ACT	GAA	GAT	2590
	Glu	Trp	Gly	Lys	Glu	Ile	Gly	Trp	Ile	Tyr	Gly	Ser	Val	Thr	Glu	Asp	
		770					775					780					
	ATC	CTG	ACG	GGT	TTC	AAG	ATG	CAT	TGC	CAT	GGA	TGG	AGA	TCT	GTG	TAC	2638
10	Ile	Leu	Thr	Gly	Phe	Lys	Met	His	Сув	His	Gly	Trp	Arg	Ser	Val	Tyr	
	785					790					795					800	
	TGT	ATG	CCT	AAG	CGT	GCA	GCT	TTT	AAA	GGA	TCT	GCT	CCT	ATT	AAC	TTG	2686
	Cys	Met	Pro	Lys	Arg	Ala	Ala	Phe	Lys	Gly	Ser	Ala	Pro	Ile	Asn	Leu	
15					805					810					815		
	TCA	GAT	CGT	CTT	CAT	CAA	GTT	CTA	CGT	TGG	GCT	CTT	GGC	TCT	GTA	GAG	2734
	Ser	Asp	Arg	Leu	His	Gln	۷al	Leu	Arg	Trp	Ala	Leu	Gly	Ser	Val	Glu	
				820					825					830			
20																	
	ATT	TTC	TTG	AGC	AGA	CAT	TGT	CCG	ATA	TGG	TAT	GGT	TAT	GGT	GGT	GGT	2782
	Ile	Phe	Leu	Ser	Arg	His	Сув	Pro	Ile	Trp	Tyr	Gly	Tyr	Gly	Gly	Gly	
			835					840					845				
25	TTA	AAA	TGG	TTG	GAG	AGA	TTC	TCT	TAC	ATC	AAC	TCT	GTC	GTC	TAT	CCT	2830
	Leu	Lys	Trp	Leu	Glu	Arg	Phe	Ser	Tyr	Ile	Asn	Ser	Val	Val	Tyr	Pro	
		850					855					860					
	TGG	ACT	TCA	CTT	CCA	TTG	ATC	GTC	TAT	TGT	TCT	CTC	ccc	GCG	GTT	TGT	2878
30	Trp	Thr	Ser	Leu	Pro	Leu	Ile	Val	Tyr	Cys	Ser	Leu	Pro	Ala	Val	Сув	
	865					870					875					880	
	TTA	CTC	ACA	GGA	AAA	TTC	ATC	GTC	CCT	GAG	ATA	AGC	AAC	TAC	GCA	GGT	2926
	Leu	Leu	Thr	Gly	Lys	Phe	Ile	Val	Pro	Glu	Ile	Ser	Asn	Tyr	Ala	Gly	
35					885					890				_	895	-	
	ATA	CTC	TTC	ATG	CTC	ATG	TTC	ATA	TCC	ATA	GCA	GTA	ACT	GGA	ATC	CTC	2974
	Ile	Leu	Phe	Met	Leu	Met	Phe	Ile	Ser	Ile	Ala	Val	Thr	Gly	Ile	Leu	
				900					905					910			
40																	

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	GAA	ATG	CAA	TGG	GGA	GGT	GTC	GGA	ATC	GAT	GAT	TGG	TGG	AGA	AAC	GAG	3022
	Glu	Met	Gln	Trp	Gly	Gly	Val	Gly	Ile	qaA	Asp	Trp	Trp	Arg	Asn	Glu	
			915					920					925				
_																	
5	CAG	TTT	TGG	GTA	ATC	GGA	GGG	GCC	TCC	TCG	CAT	CTA	TTT	GCT	CTG	TTT	3070
	Gln	Phe	Trp	Val	Ile	Gly	Gly	Ala	Ser	Ser	His	Leu	Phe	Ala	Leu	Phe	
		930					935					940					
10		-		-	AAA												3118
10		Gly	Leu	Leu	Lys		Leu	Ala	GIA	Val		Thr	Asn	Phe	Thr		
	945					950					955					960	
	л ст	TCA	מממ	CCA	GCA	CAC	CAT	GGA	COT	ሙሞር	<b></b>	GVG	CTT	TAC	<b>ል</b> ሞር	<del>ተተ</del> ር	3166
					Ala												3100
15	1111	561	Lys	nia	965	nop	nap	U1,	710	970	501	<b>01</b> u	acu	.,.	975	7110	
• •					703					,,,							
	AAG	TGG	ACA	ACT	TTG	TTG	ATT	CCT	CCG	ACA	ACA	СТТ	CTG	ATC	ATT	AAC	3214
					Leu												
	-1-			980					985		••			990			
20																	
	ATC	ATT	GGA	GTT	ATT	GTC	GGC	GTT	TCT	GAT	GCC	ATT	AGC	AAT	GGC	TAT	3262
	Ile	Ile	Gly	Val	Ile	Val	Gly	Val	Ser	Asp	Ala	Ile	Ser	Asn	Gly	Tyr	
			995					1000	0				100	5			
25	GAC	TCA	TGG	GGA	CCT	CTC	TTT	GGG	AGA	CTT	TTC	TTC	GCT	CTT	TGG	GTC	3310
	Asp	Ser	Trp	Gly	Pro	Leu	Phe	Gly	Arg	Leu	Phe	Phe	Ala	Leu	Trp	Val	
		1010	)				1019	5				1026	)				
					TAC												3358
30	Ile	Val	His	Leu	Tyr	Pro	Phe	Leu	ГÀв	Gly	Met	Leu	Gly	Lys	Gln	Asp	
	102	5				1030	0				1039	5				1040	
					ATT												3406
35	Lys	Met	Pro	Thr	Ile		Val	Val	Trp			Leu	Leu	Ala			
ננ					1049	•				1050	J				1059	•	
	تكليك	ארא	CTC	danica danica	TGG	GTC.	አርማ	N electr	220	CCC	drama	CI TOVO	~~~	***	CCC	CCA	7454
																	3454
	neu	TILL	neu	1060	Trp	val	wrd	rre	106		rne	AST	AIG	1076	-	GIÀ	
				1000	•				100	•				10/0	•		

WO 98/00549	PCT/AU97/00402

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	CCA GTG TTG GAG ATC TGT GGT CTG AAT TGT GGA AAC TAAGATCCTC	3500
	Pro Val Leu Glu Ile Cys Gly Leu Asn Cys Gly Asn	
	1075 1080	
5	AGTGAAAGAA GAGCAAAGGA GTTTGTGTTG GAGCTTTGGA AGCAAATGTG TTGATGATGA	3560
	TGCAAGTGTG TTTGTAGACA AAGATGTGCA GTTTTTACTT TTTACGACTT GTTAAACCTT	2620
	ISCARGIGIG TITGTAGACA AAGATGIGCA GITTITACIT TITACGACTI GITAAACCIT	3620
	TTTTGTTACC CCTAAATTAA TTCTTTTGTT ATCATGGTTA TACTAATAGA ATTGTTTGTT	3680
10		
	TTTCTTTTTT ACATGTACTT TTAGTTATTC CGTAGTTATT GTATAATACT GATAACGATC	3740
	ATATATACAC ACTITGITAA CAAAAAAAAA AAAAAAAAA AAAAAAAAA AAAGCGGCCG	3800
15	CTCGAATTGT CGACGCGGCC GCGAATTC	3828
20	(2) 7372234 722 722 72 32	
20	(2) INFORMATION FOR SEQ ID NO:8:	
	(i) SEQUENCE CHARACTERISTICS:	
	(A) LENGTH: 1084 amino acids	
	(B) TYPE: amino acid	
25	(D) TOPOLOGY: linear	
	(ii) MOLECULE TYPE: protein	
	•	
	(xi) SEQUENCE DESCRIPTION: SEQ ID NO:8:	
30	(AL) DESCRICE SESCRETIZATE SES ID NO. 0.	
50		
	Met Asn Thr Gly Gly Arg Leu Ile Ala Gly Ser His Asn Arg Asn Glu	
	1 5 10 15	
	Phe Val Leu Ile Asn Ala Asp Glu Ser Ala Arg Ile Arg Ser Val Gln	
35	20 25 30	
	Glu Leu Ser Gly Gln Thr Cys Gln Ile Cys Gly Asp Glu Ile Glu Leu	
	35 40 45	

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	Thr	Val	Ser	Ser	Glu	Leu	Phe	Val	Ala	Cys	Asn	Glu	Cha	Ala	Phe	Pro
		50					55					60				
	Val	Сув	Arg	Pro	Сув	Tyr	Glu	Tyr	Glu	Arg	Arg	Glu	Gly	Asn	Gln	Ala
5	65					70					75					80
	Cys	Pro	Gln	Cys	Lys	Thr	Arg	Tyr	Lys	Arg	Ile	Lys	Gly	Ser	Pro	Arg
					85					90					95	
10	Val	Asp	Gly	Asp	Asp	Glu	Glu	Glu	Glu	Asp	Ile	Asp	Asp	Leu	Glu	Tyr
				100					105					110		
	Glu	Phe	Asp	His	Gly	Met	Asp	Pro	Glu	His	Ala	Ala	Glu	Ala	Ala	Leu
			115					120					125			
15																
	Ser	Ser	Arg	Leu	Asn	Thr	Gly	Arg	Gly	Gly	Leu	Asp	Ser	Ala	Pro	Pro
		130					135					140				
20	Gly 145	Ser	Gln	Ile	Pro		Leu	Thr	Tyr	Сув		Glu	Asp	Ala	Asp	
20	143					150					155					160
20		°02	Nan	N ===	ui o		, on	T) o	Val	Dwa		Com	mb	<b>63.</b> 4	Th ess	
20		Ser	Asp	Arg			Leu	Ile	Val			Ser	Thr	Gly	Tyr	
20		Ser	Asp	Arg	His 165		Leu	Ile	Val	Pro 170		Ser	Thr	Gly	Tyr 175	
	Tyr				165	Ala				170	Pro				175	Gly
				Tyr	165	Ala			Thr	170	Pro			Pro	175	Gly
	Tyr				165	Ala				170	Pro				175	Gly
	Tyr	Arg	Val	Tyr 180	165 Pro	Ala Ala	Pro	Phe	Thr 185	170 Asp	Pro Ser	Ser	Ala	Pro 190	175	Gly Gln
	Tyr	Arg	Val	Tyr 180	165 Pro	Ala Ala	Pro	Phe	Thr 185	170 Asp	Pro Ser	Ser	Ala	Pro 190	175 Pro	Gly Gln
	Tyr	Arg	Val Ser	Tyr 180	165 Pro	Ala Ala	Pro	Phe Lys	Thr 185	170 Asp	Pro Ser	Ser	Ala Tyr	Pro 190	175 Pro	Gly Gln
25	Tyr Asn Ala	Arg Arg	Val Ser 195	Tyr 180 Met	165 Pro Val	Ala Ala Pro	Pro Gln	Phe Lys 200	Thr 185 Asp	170 Asp	Pro Ser	Ser Glu	Ala Tyr 205	Pro 190 Gly	175 Pro	Gly Gln Gly
25	Tyr Asn Ala	Arg Arg	Val Ser 195	Tyr 180 Met	165 Pro Val	Ala Ala Pro	Pro Gln	Phe Lys 200	Thr 185 Asp	170 Asp	Pro Ser	Ser Glu	Ala Tyr 205	Pro 190 Gly	175 Pro	Gly Gln Gly
25	Tyr Asn Ala	Arg Arg Val	Val Ser 195	Tyr 180 Met	165 Pro Val	Ala Ala Pro	Pro Gln Arg	Phe Lys 200	Thr 185 Asp	170 Asp	Pro Ser	Ser Glu Lys	Ala Tyr 205	Pro 190 Gly	175 Pro	Gly Gln Gly
25	Tyr Asn Ala	Arg Arg Val 210	Val Ser 195	Tyr 180 Met	Pro Val	Ala Ala Pro	Pro Gln Arg 215	Phe Lys 200 Met	Thr 185 Asp Glu	170 Asp Ile Val	Pro Ser Ala	Ser Glu Lys 220	Ala Tyr 205 Arg	Pro 190 Gly	175 Pro	Gly Gln Gly
25 30	Tyr Asn Ala	Arg Arg Val 210	Val Ser 195	Tyr 180 Met	Pro Val	Ala Ala Pro	Pro Gln Arg 215	Phe Lys 200 Met	Thr 185 Asp Glu	170 Asp Ile Val	Pro Ser Ala	Ser Glu Lys 220	Ala Tyr 205 Arg	Pro 190 Gly	175 Pro Tyr	Gly Gln Gly
25 30	Tyr Asn Ala Ser	Arg Arg Val 210	Val Ser 195	Tyr 180 Met	Pro Val	Ala Ala Pro Asp	Pro Gln Arg 215	Phe Lys 200 Met	Thr 185 Asp Glu	170 Asp Ile Val	Pro Ser Ala Trp	Ser Glu Lys 220	Ala Tyr 205 Arg	Pro 190 Gly	175 Pro Tyr	Gly Gln Gly
25 30	Tyr Asn Ala Ser Glu 225	Arg Val 210 Lys	Val Ser 195 Ala	Tyr 180 Met Trp	165 Pro Val Lys	Ala Pro Asp Ile 230	Pro Gln Arg 215 Lys	Phe Lys 200 Met	Thr 185 Asp Glu	170 Asp Ile Val	Pro Ser Ala Trp Gly 235	Ser Glu Lys 220 Asn	Ala Tyr 205 Arg	Pro 190 Gly Arg	175 Pro Tyr	Gly Gly Gly 240

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	Glu	Gly	Arg	Gln	Pro	Leu	Ser	Arg	Lys	Leu	Pro	Ile	Arg	Ser	Ser	Arg
				260					265					270		
	Ile	Asn	Pro	Tyr	Arg	Met	Leu	Ile	Leu	Cys	Arg	Leu	Ala	Ile	Leu	Gly
5			275					280					285			
	Leu	Phe	Phe	His	Tyr	Arg	Ile	Leu	His	Pro	Val	Asn	Asp	Ala	Tyr	Gly
		290					295					300				
10																
10	Leu	Trp	Leu	Thr	Ser	Val	Ile	Сув	Glu	Ile	Trp	Phe	Ala	Val	Ser	Trp
	305					310					315					320
	Ile	Leu	Asp	Gln		Pro	Lys	Trp	Tyr		Ile	Glu	Arg	Glu	Thr	Tyr
1.5					325					330					335	
15																
	Leu	Asp	Arg		Ser	Leu	Arg	Tyr		Lys	Glu	Gly	Lys		Ser	Gly
				340					345					350		
	_		_				-,		_	_,		_	_			
20	Leu	Ala		vai	Asp	Val	Pne		ser	Thr	vaı	Asp		Leu	Lys	Glu
20			355					360					365			
	Dro	Dro	T an	T) a	The	Ala	han	The	Ma l	T 011	Com	T1.	T	21-	V-1	<b>&gt;</b>
	PIO	370	Leu	116	1111	AIG	375	1111	vai	TEU	ser	380	Leu	Ala	vaı	Авр
		370					313					300				
25	туг	Pro	Val	ap A	Lvs	Val	Δla	Cva	Tvr	Val	Ser	Aan	Ann	G1v	ב [ ת	פות
	385				2,0	390	,,,,	Cyo	-1-	***	395	A9II	Noti	Gry	A.a	400
																100
	Met	Leu	Thr	Phe	Glu	Ala	Leu	Ser	Asp	Thr	Ala	asa	Phe	Ala	Thr	lvs
					405					410					415	_,_
30																
	Trp	Val	Pro	Phe	Cys	Lys	Lys	Phe	Asn	Ile	Glu	Pro	Arg	Ala	Pro	Glu
	=			420	•	•	•		425					430		
	Trp	Tyr	Phe	Ser	Gln	Lys	Met	Asp	Tyr	Leu	Lys	Asn	Lvs	Val	His	Pro
35	-	•	435			•		440	•		•		445			
	Ala	Phe	Val	Arg	Glu	Arg	Arg	Ala	Met	Lys	Arg	Asp	Tyr	Glu	Glu	Phe
		450					455				_	460	•			

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	Lys 465	Val	rys	Ile	Asn	Ala 470	Leu	Val	Ala	Thr	Ala 475	Gln	Lys	Val	Pro	G1:
	G) u	Ara	Trn	Thr	Mat	Gln	Aen	Gly	Thr	Pro	Tvv	Pro	C) v	Nan	Aon	Val
5	Giu	My	p	1111	485	GIII	vah	GIY	1111	490	11p	PIO	GIY	Abn	495	val
	Arq	Asp	His	Pro	Glv	Met	Ile	Gln	Val	Phe	Leu	Glv	His	Ser	Glv	Val
	,	•		500	•				505			2		510	,	
10	Arg	Asp	Thr	Asp	Gly	Asn	Glu	Leu	Pro	Arg	Leu	Val	Tyr	Val	Ser	Arg
			515					520					525			
	Glu	Lys	Arg	Pro	Gly	Phe	Asp	His	His	Lys	Lys	Ala	Gly	Ala	Met	Asn
15		530					535					540				
		Leu	Ile	Arg	Val		Ala	Val	Leu	Ser		Ala	Pro	Tyr	Leu	
	545					550					555					560
20	Asn	Val	Asp	Сув	Asp 565	His	Tyr	Ile	Asn	Asn 570	Ser	Lys	Ala	Ile	_	Glu
20					262					570					575	
	Ser	Met	Cys	Phe 580	Met	Met	Asp	Pro	Gln 585	Ser	Gly	Lys	Lys	Val 590	Сув	Туг
25																
25	Val	Gln	Phe 595	Pro	Gln	Arg	Phe	Asp	Gly	Ile	Asp	Arg	His 605	Asp	Arg	Tyr
								-1			_				_	
	ser	610	Arg	Asn	vai	vaı	615	Pne	Asp	116	Asn	Met 620	Lys	GIA	Leu	Asp
30	Glv	Ile	Gln	Gly	Pro	Ile	Tvr	Va1	Glv	Thr	Glv	Cva	Val	Dhe	Ara	Lve
	625			,		630	•,,•	Vu.2	Oly	****	635	Cys	vai	FIIC	Arg	640
	Gln	Ala	Leu	Tyr	Gly	Phe	Asp	Ala	Pro	Lya	Lys	Lys	Lys	Pro	Pro	Gly
35					645					650	-	-	-		655	•
	Lys	Thr	Суз	Asn	Cys	Trp	Pro	Lys	Trp	Сув	Сув	Leu	Сув	Сув	Gly	Leu
				660					665					670		

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	Arg	Lys	Lys 675	Ser	Lys	Thr	Lys	Ala 680	Thr	Asp	Lys	Lys	Thr 685	Asn	Thr	Lys
5	Glu	Thr 690	Ser	Lys	Gln	Ile	His 695	Ala	Leu	Glu	Asn	Val 700	Asp	Glu	Gly	Val
	Ile 705	Val	Pro	Val	Ser	Asn 710	Val	Glu	Lys	Arg	Ser 715	Glu	Ala	Thr	Gln	Leu 720
10	Lys	Leu	Glu	Lys	Lys 725	Phe	Gly	Gln	Ser	Pro 730	Val	Phe	Val	Ala	Ser 735	Ala
15	Val	Leu	Gln	Asn 740	Gly	Gly	Val	Pro	Arg 745	Asn	Ala	Ser	Pro	Ala 750	Сув	Leu
.,	Leu	Arg	Glu 755	Ala	Ile	Gln	Val	Ile 760	Ser	Сув	Gly	Tyr	Gln 765	Asp	Lys	Thr
20	Glu	Trp 770	Gly	Lys	Glu	Ile	Gly 775	Trp	Ile	Tyr	Gly	Ser 780	Val	Thr	Glu	Asp
	Ile 785	Leu	Thr	Gly	Phe	Lys 790	Met	His	Сув	His	Gly 795	Trp	Arg	Ser	Val	Tyr 800
25	Сув	Met	Pro	Lys	Arg 805	Ala	Ala	Phe	Lys	Gly 810	Ser	Ala	Pro	Ile	Asn 815	Leu
30	Ser	Asp	Arg	Leu 820	Hıs	Gln	Val	Leu	Arg 825	Trp	Ala	Leu	Gly	Ser 830	Val	Glu
JU	Ile	Phe	Leu 835	Ser	Arg	Нів	Cys	Pro 840	Ile	Trp	Tyr	Gly	Tyr 845	Gly	Gly	Gly
35	Leu	Lys 850	Trp	Leu	Glu	Arg	Phe 855	Ser	Tyr	Ile	Asn	Ser 860	Val	Val	Туг	Pro
	Trp 865	Thr	Ser	Leu	Pro	Leu 870	Ile	Val	Tyr	Суз	Ser 875	Leu	Pro	Ala	Val	Cys

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	Leu	Leu	Thr	Gly	Lys 885	Phe	Ile	Val	Pro	Glu 890	Ile	Ser	Asn	Tyr	Ala 895	Gly
5	Ile	Leu	Phe	Met 900	Leu	Met	Phe	Ile	Ser 905	Ile	Ala	Val	Thr	Gly 910	Ile	Leu
	Glu	Met	Gln 915	Trp	Gly	Gly	Val	Gly 920	Ile	Asp	Asp	Trp	Trp 925	Arg	Asn	Glu
10	Gln	Phe 930	Trp	Val	Ile	Gly	Gly 935	Ala	Ser	Ser	His	Leu 940	Phe	Ala	Leu	Phe
15	Gln 945	Gly	Leu	Leu	Lys	Val 950	Leu	Ala	Gly	Val	Asn 955	Thr	Asn	Phe	Thr	Val 960
•	Thr	Ser	Lys	Ala	Ala 965	Asp	Авр	Gly	Ala	Phe 970	Ser	Glu	Leu	Туг	Ile 975	Phe
20	Lys	Trp	Thr	Thr 980	Leu	Leu	Ile	Pro	Pro 985	Thr	Thr	Leu	Leu	Ile 990	Ile	Asn
	Ile	Ile	Gly 995	Val	Ile	Val	Gly	Val		Asp	Ala	Ile	Ser		Gly	Tyr
25	Asp	Ser		Gly	Pro	Leu	Phe 1019		Arg	Leu	Phe	Phe 1020		Leu	Trp	Val
30	Ile 1025		His	Leu	Tyr	Pro 1030		Leu	Lys	Gly	Met 1035		Gly	Lys	Gln	Asp 1040
30	Lys	Met	Pro	Thr	Ile 1045		Val	Val	Trp	Ser 1050		Leu	Leu	Ala	Ser 1055	
35	Leu	Thr	Leu	Leu 1060	_	Val	Arg	Ile	Asn 1065		Phe	Val	Ala	Lys 1070	-	Gly
	Pro	Val	Leu 1079	Glu 5	Ile	Сув	Gly	Leu 1080		Сув	Gly	Asn				

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(2) INFORMATION FOR SEQ ID NO:9:

	(i) SEQUENCE CHARACTERISTICS:	
	(A) LENGTH: 3614 base pairs	
5	(B) TYPE: nucleic acid	
	(C) STRANDEDNESS: single	
	(D) TOPOLOGY: linear	
	(ii) MOLECULE TYPE: cDNA	
10		
	(iii) HYPOTHETICAL: NO	
	(vi) ORIGINAL SOURCE:	
	(A) ORGANISM: Arabidopsis thaliana	
15	(B) STRAIN: Columbia	
	(vii) IMMEDIATE SOURCE:	
	(B) CLONE: Ath-B	
20		
20	(ix) FEATURE:	
	(A) NAME/KEY: CDS	
	(B) LOCATION: 2173411	
25	(with applicable production and to up a	
23	(xi) SEQUENCE DESCRIPTION: SEQ ID NO:9:	
	GAATTCGCGG CCGCGTCGAC TACGGCTGCG AGAAGACGAC AGAAGGGGGAT CCCAAGATTC	60
	GANTICOCOO CCOCOTCOAC TACOOCTOCO AOAAGACGAC AGAAGGGGAT CCCAAGATTC	01
	TCCTCTTCGT CTTCCTTATA AACTATCTCT CTGTAGAGAA GAAAGCTTGG ATCCAGATTG	120
30		-20
	AGAGAGATTC AGAGAGCCAC ATCACCACAC TCCATCTTCA GATCTCATGA TTTGAACTAT	180
	TCCGACGTTT CGGTGTTGGA AGCAACTAAG TGACAA ATG GAA TCC GAA GGA GAA	234
	Met Glu Ser Glu Gly Glu	
35	1 5	
	ACC GCG GGA AAG CCG ATG AAG AAC ATT GTT CCG CAG ACT TGC CAG ATC	282
	Thr Ala Gly Lys Pro Met Lys Asn Ile Val Pro Gln Thr Cys Gln Ile	
	10 15 20	
<b>4</b> 0		

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	TGT	AGT	GAC	AAT	GTT	GGC	AAG	ACT	GTT	GAT	GGA	GAT	CGT	TTT	GTG	GCT	330
	Сув	Ser	Asp	Asn	Val	Gly	Lys	Thr	Val	Авр	Gly	qeA	Arg	Phe	Val	Ala	
			25					30					35				
_																	
5	TGT	GAT	ATT	TGT	TCA	TTC	CCA	GTT	TGT	CGG	CCT	TGC	TAC	GAG	TAT	GAG	378
	Cys	-	Ile	Cys	Ser	Phe		Val	Cys	Arg	Pro	_	Tyr	Glu	Tyr	Glu	
		40					45					50					
	3.00		Chm	ccc	አስጥ	CD D	w-m	mar.		CNC	<b>7</b> 00		200	202	m> c	***	425
10				GGG Gly													426
10	55	Буз	voh	Gly	ASII	60	361	Cys	FIO	GIII	65	Був	1111	Arg	ıyı	70	
	,,,					00					03					,,	
	AGG	CTC	AAA	GGT	AGT	CCT	GCT	ATT	CCT	GGT	GAT	AAA	GAC	GAG	GAT	GGC	474
				Gly													
15	Ĭ		•	Ī	75					80	•	•	•		85	•	
	TTA	GCT	GAT	GAA	GGT	ACT	GTT	GAG	TTC	AAC	TAC	CCT	CAG	AAG	GAG	AAA	522
	Leu	Ala	Asp	Glu	Gly	Thr	Val	Glu	Phe	Asn	Tyr	Pro	Gln	Lys	Glu	Lys	
				90					95					100			
20																	
	ATT	TCA	GAG	CGG	ATG	CTT	GGT	TGG	CAT	CTT	ACT	CGT	GGG	AAG	GGA	GAG	570
	Ile	Ser	Glu	Arg	Met	Leu	Gly	Trp	His	Leu	Thr	Arg	Gly	Lys	Gly	Glu	
			105					110					115				
0.5																	
25				GAA													618
	Glu		Gly	Glu	Pro	Gln	_	Ąsp	Lys	Glu	Val		His	neA	His	Leu	
		120					125					130					
	CCT	CCT	CTC	ACG	200	707	CAA	CAT	n cam	mc»	CON	C3.0	mmm	mom	a com	000	
30				Thr													666
50	135	AL 9	Deu	****	561	140	<b>J1</b> 11	nop	1111	261	145	GIU	FIIC	261	VIG	150	
																130	
	TCA	CCT	GAA	CGC	CTC	TCT	GTA	TCT	тст	ACT	ATC	GCT	GGG	GGA	AAG	CGC	714
				Arg													
35					155					160			-	_	165		
	CTT	CCC	TAT	TCA	TCA	GAT	GTC	AAT	CAA	TCA	CCA	AAT	AGA	AGG	ATT	GTG	762
	Leu	Pro	Tyr	Ser	Ser	Asp	Val	Asn	Gln	Ser	Pro	Asn	Arg	Arg	Ile	Val	
				170					175					180			
40																	

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	GAT	CCT	GTT	GGA	CTC	GGG	AAT	GTA	GCT	TGG	AAG	GAG	AGA	GTT	GAT	GGC	810
	Asp	Pro	Val	Gly	Leu	Gly	Asn	Val	Ala	Trp	Lys	Glu	Arg	Val	Asp	Gly	
			185					190					195				
_																	
5	TGG	AAA	ATG	AAG	CAA	GAG	AAG	AAT	ACT	GGT	CCT	GTC	AGC	ACG	CAG	GCT	858
	Trp	Lys	Met	Lys	Gln	Glu	Lys	Asn	Thr	Gly	Pro	Val	Ser	Thr	Gln	Ala	
		200					205					210					
. ^					GGT												906
IU		Ser	Glu	Arg	Gly		Val	Asp	Ile	Asp		Ser	Thr	Asp	Ile	Leu	
	215					220					225					230	
	<b></b>	a.m	a. a		c.m.c.	omo											
					CTG												954
15	AIA	Авр	Giu	ATA	Leu	Leu	Asn	Asp	Glu		Arg	GIn	Leu	Leu		Arg	
כו					235					240					245		
		CTT	TC N	s mar	CCT	TC N	TCA	cca	N.T.O.	N N TT	CCT	m».c	202	3 mc	omm.	3 mm	1000
					Pro												1002
	цуъ	vai	361	250	PIO	361	361	Arg	255	ASII	PLO	TAL	Arg	260	Val	116	
20				250					233					260			
	ΔTG	CTG	ccc	СТТ	GTT	ልጥሮ	CTT	тст	CTC	<b>ጥ</b> ጉር	TTC	ሮልጥ	<b>ፐ</b> እር	CGT	እጥአ	מכמ	1050
					Val												1030
			265	200	Vu.	110	Deu	270	200		Deu	1120	275	ALY	116	****	
			203					2,0					2,5				
25	AAC	CCA	GTG	CCA	AAT	GCC	TTT	GCT	CTA	TGG	CTG	GTC	TCT	GTG	ATA	TGT	1098
					Asn												
		280					285			•		290				•	
	GAG	ATC	TGG	TTT	GCC	TTA	TCC	TGG	ATT	TTG	GAT	CAG	TTT	CCC	AAG	TGG	1146
30	Glu	Ile	Trp	Phe	Ala	Leu	Ser	Trp	Ile	Leu	Asp	Gln	Phe	Pro	Lys	Trp	
	295					300					305					310	
	TTT	CCT	GTG	AAC	CGT	GAA	ACC	TAC	CTC	GAC	AGG	CTT	GCT	TTA	AGA	TAT	1194
	Phe	Pro	Val	Asn	Arg	Glu	Thr	Tyr	Leu	Asp	Arg	Leu	Ala	Leu	Arg	Tyr	
35					315					320					325		
	GAT	CGT	GAA	GGT	GAG	CCA	TCA	CAG	TTA	GCT	GCT	GTT	GAC	ATT	TTC	GTG	1242
	Asp	Arg	Glu	Gly	Glu	Pro	Ser	Gln	Leu	Ala	Ala	Val	Asp	Ile	Phe	Val	
				330					335					340			

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	AGT	ACT	GTT	GAC	CCC	TTG	AAG	GAG	CCA	CCC	CTT	GTG	ACA	GCC	AAC	ACA	1290
	Ser	Thr	Val	Asp	Pro	Leu	Lys	Glu	Pro	Pro	Leu	Val	Thr	Ala	Asn	Thr	
			345					350					355				
5	GTG	CTC	TCT	ATT	CTG	GCT	GTT	GAC	TAC	CCA	GTT	GAC	AAG	GTG	TCC	TGT	1338
	Val	Leu	Ser	Ile	Leu	Ala	Val	Авр	Tyr	Pro	Val	Asp	Lys	Val	Ser	Сув	
		360					365					370					
	TAT	GTT	TCT	GAT	GAT	GGT	GCT	GCT	ATG	TTA	TCA	TTT	GAA	TÇA	CTT	GCA	1386
10	Tyr	Val	Ser	Asp	Asp	Gly	Ala	Ala	Met	Leu	Ser	Phe	Glu	Ser	Leu	Ala	
	375					380					385					390	
				GAG													1434
1.5	Glu	Thr	Ser	Glu		Ala	Arg	ГÀв	Trp		Pro	Phe	Cys	Lys	-	Tyr	
15					395					400					405		
		_															
				CCT							"						1482
	Ser	He	Glu	Pro	Arg	Ala	Pro	Glu	_	Tyr	Phe	Ala	Ala	-	He	Asp	
20				410					415					420			
20	Th C	ww.	220	CAT	222	- Comm	CNC	202	TC N	marin	OTC.		C N TO	~~~	202	COTT	1530
				GAT													1530
	171	neu	-	Asp	гуя	val	GIN	430	ser	Pne	vai	гåя	_	Arg	Arg	Ala	
			425					430					435				
25	ΔTG	DAG	≱GG	GAA	<b>יי</b> מיי	GAG	CAA	John	מממ	ልሞር	CCIA	ልሞር	ידממ	CCA	سس	CTT	1578
				Glu													1370
		440	***- 5		-7-	014	445		2,0		****	450	7,011	744	DCu	742	
							115					130					
	TCC	AAA	GCC	CTA	AAA	TGT	CCT	GAA	GAA	GGG	TGG	GTT	ATG	CAA	GAT	GGC	1626
30				Leu													
	455	•			•	460					465					470	
	ACA	CCG	TGG	CCT	GGA	AAT	AAT	ACA	GGG	GAC	CAT	CCA	GGA	ATG	ATC	CAG	1674
	Thr	Pro	Trp	Pro	Gly	Asn	Asn	Thr	Gly	Asp	His	Pro	Gly	Met	Ile	Gln	
35					475					480			_		485		
	GTC	TTC	TTA	GGG	CAA	AAT	GGT	GGA	CTT	GAT	GCA	GAG	GGC	AAT	GAG	CTC	1722
	Val	Phe	Leu	Gly	Gln	Asn	Gly	Gly	Leu	Asp	Ala	Glu	Gly	Asn	Glu	Leu	
				490					495					500			
40																	

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	CCG	CGT	TTG	GTA	TAT	GTT	тст	CGA	GAA	AAG	CGA	CCA	GGA	TTC	CAG	CAC	1770
	Pro	Arg	Leu	Val	Tyr	Val	Ser	Arg	Glu	Lys	Arg	Pro	Gly	Phe	Gln	His	
			505					510					515				
_																	
3					GGT												1818
	His	-	Lys	Ala	Gly	Ala		Asn	Ala	Leu	Val	•	Val	Ser	Ala	Val	
		520					525					530					
	ىلىلى	ልሮሮ	አልጥ	CGA	CCT	ጥጥር	ልሞሮ	ምጥር	ልልጥ	ርጥጥ	CAT	TYTT	СУТ	CDT	ሞአር	እጥክ	1866
10					Pro												1000
	535		7.01.	O.,	710	540		LCu	AOII	Deu	545	cys	veħ	nis	TYL	550	
	777					J.,					313					330	
	AAT	AAC	AGC	AAA	GCC	TTA	AGA	GAA	GCA	ATG	TGC	TTC	стс	ATG	GAC	CCA	1914
					Ala												
15				•	555		•			560	•				565		
	AAC	CTC	GGG	AAG	CAA	GTT	TGT	TAT	GTT	CAG	TTC	CCA	CAA	AGA	TTT	GAT	1962
	Asn	Leu	Gly	Lys	Gln	Val	Cys	Tyr	Val	Gln	Phe	Pro	Gln	Arg	Phe	Asp	
				570					575					580			
20																	
	GGT	ATC	GAT	AAG	AAC	GAT	AGA	TAT	GCT	AAT	CGT	AAT	ACC	GTG	TTC	TTT	2010
	Gly	Ile	Asp	Lys	Asn	Asp	Arg	Tyr	Ala	Asn	Arg	Asn	Thr	Val	Phe	Phe	
			585					590					595				
25	GAT	ATT	AAC	TTG	AGA	GGT	TTA	GAT	GGG	ATT	CAA	GGA	CCT	GTA	TAT	GTC	2058
	Asp	Ile	Asn	Leu	Arg	Gly	Leu	Asp	Gly	Ile	Gln	Gly	Pro	Val	Tyr	Val	
		600					605					610					
30					GTT												2106
30		Thr	GIY	Cys	Val		Asn	Arg	Thr	Ala		Tyr	Gly	Tyr	Glu		
	615					620					625					630	
	CCD	እመአ	222	OW.		<b>C2.</b> C			001	1 Cm	omm.						
					AAA												2154
35	PLO	116	ьув	val	Lys 635	บรร	Lys	гув	PIO	640	Leu	Leu	ser	Lys	645	Сув	
					033					040					043		
	GGT	GGA	TCA	AGA	AAG	AAG	дат	TCC	ааа	GCT	AAG	ддд	GAG	TCG	GAC	AAA	2202
					Lys												
	•	-		650	_				655		•	•		660		-4 -	
40																	

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	AAG	AAA	TCA	GGC	AGG	CAT	ACT	GAC	TCA	ACT	GTT	CCT	GTA	TTC	AAC	CTC	2250
	Lys	Lys	Ser	Gly	Arg	His	Thr	Asp	Ser	Thr	Val	Pro	Val	Phe	Asn	Leu	
			665					670					675				
_																	
5					GAG					_					_		2298
	Asp	-	Ile	Glu	Glu	Gly		Glu	Gly	Ala	Gly		Asp	qeA	Glu	Lys	
		680					685					690					
	000	CTC	ጥጥአ	איזעיי	TCG	CAA	n TYC	NGC.	Calac	GNG	D D C	CCA	Դուր	CCA	CNG	ጥርጥ	2346
10					Ser												2340
	695	DCu	200	.,	501	700	1,00	552			705			,		710	
	***																
	GCT	GTT	TTT	GTT	GCT	TCT	ACC	CTA	ATG	GAA	AAT	GGT	GGT	GTT	CCT	CCT	2394
	Ala	Val	Phe	Val	Ala	Ser	Thr	Leu	Met	Glu	Asn	Gly	Gly	Val	Pro	Pro	
15					715					720					725		
	TCA	GCA	ACT	CCA	GAA	AAC	TTT	CTC	AAA	GAG	GCT	ATC	CAT	GTC	ATT	AGT	2442
	Ser	Ala	Thr	Pro	Glu	Asn	Phe	Leu	Lys	Glu	Ala	Ile	His	Val	Ile	Ser	
				730					735					740			
20																	
					GAT												2490
	Сув	Gly		Glu	Asp	Lys	Ser		Trp	Gly	Met	Glu		Gly	Trp	Ile	
			745					750					755				
25	ጥልጥ	GGT	ጥርጥ	GTG	ACA	GAA	тар	<b>ስ</b> ጥጥ	CTG	ΔСТ	GGG	ጥጥር	444	ATG	САТ	GCC	2538
					Thr												
	-,-	760				•••	765			••••	,	770					
	CGT	GGA	TGG	CGA	TCC	ATT	TAC	TGC	ATG	CCT	AAG	CTT	CCA	GCT	TTC	AAG	2586
30	Arg	Gly	Trp	Arg	Ser	Ile	Tyr	Cys	Met	Pro	Lys	Leu	Pro	Ala	Phe	Lys	
	775					780					785					790	
	GGT	TCT	GCT	CCT	ATC	AAT	CTT	TCA	GAT	CGT	CTG	AAC	CAA	GTG	CTG	AGG	2634
	Gly	Ser	Ala	Pro	Ile	Asn	Leu	Ser	Asp	Arg	Leu	Asn	Gln	Val	Leu	Arg	
35					795					800					805		
		_											<b>.</b>				
					TCA												2682
	Trp	Ala	Leu	_	Ser	val	Glu	116		PUE	ser	Arg	H18	-	Pro	116	
				810					815					820			

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	TGG	TAT	GGT	TAC	TAA	GGG	AGG	CTA	AAA	TTT	CTT	GAG	AGG	TTT	GCG	TAT	2730
	Trp	Tyr	Gly	Tyr	Asn	Gly	Arg	Leu	Lys	Phe	Leu	Glu	Arg	Phe	Ala	Tyr	
			825					830					835				
5	GTG	AAC	ACC	ACC	ATC	TAC	CCT	ATC	ACC	TCC	TTA	CCT	CTT	CTC	ATG	TAT	2778
	Val	Asn	Thr	Thr	Ile	Tyr	Pro	Ile	Thr	Ser	Ile	Pro	Leu	Leu	Met	Tyr	
		840					845					850					
	TGT	ACA	TTG	CTA	GCC	GTT	TGT	CTC	TTC	ACC	AAC	CAG	TTT	ATT	ATT	CCT	2826
10	Cys	Thr	Leu	Leu	Ala	Val	Cys	Leu	Phe	Thr	Asn	Gln	Phe	Ile	Ile	Pro	
	855					860					865					870	
	CAG	ATT	AGT	AAC	ATT	GCA	AGT	ATA	TGG	TTT	CTG	TCT	CTC	TTT	CTC	TCC	2874
	Gln	Ile	Ser	Asn	Ile	Ala	Ser	Ile	Trp	Phe	Leu	Ser	Leu	Phe	Leu	Ser	
15					875					880					885		
						ATA											2922
	Ile	Phe	Ala		Gly	Ile	Leu	Glu		Arg	Trp	Ser	Gly		Gly	Ile	
20				890					895					900			
20									-	<b></b>	ama			~~	am.	maa	2070
																TCC	2970
	Asp	Glu	-	Trp	Arg	Asn	GIu		Phe	Trp	vai	He	_	GIY	vai	ser	
			905					910					915				
25	aam	C2 M	mma	mmo		ama	-	<i>~</i>	com	N/II/C	<b>a</b> ma		ama	<b></b>	~~~	GGT	3018
23																Gly	3016
	ALA	920		FIIC	ALG	Val	925	GIII	GIY	116	neu	930	AGI	beu	NIG	Gly	
		920					723					330					
	ልተተ	GAC	ACA	AAC	ፐጥሮ	ACA	GTT	ACC	ДОТ	ДДД	GCT	ፐርል	GAT	GAA	GAC	GGA	3066
30																Gly	3000
30	935	_	••••	• •		940				-,-	945	-		-		950	
											- ,-						
	GAC	TTT	GCT	GAG	CTC	TAC	TTG	TTC	AAA	TGG	ACA	ACA	CTT	CTG	ATT	CCG	3114
																Pro	
35	-				955	_			•	960					965		
	CCA	ACG	ACG	CTG	CTC	: ATT	GTA	AAC	TTA	GTG	GGA	GTI	GTT	GCA	GGA	GTC	3162
	Pro	Thr	Thr	Leu	Leu	Ile	Val	Asn	Leu	Val	Gly	Val	Val	Ala	Gly	Val	
				970	ı				975					980	•		
. 40																	

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	TCT	TAT	GCT	ATC	AAC	AGT	GGA	TAC	CAA	TCA	TGG	GGA	CCA	CTC	TTT	GGT	3210
	Ser	Tyr	Ala	Ile	Asn	Ser	Gly	Tyr	Gln	Ser	Trp	Gly	Pro	Leu	Phe	Gly	
			985					990					995				
_																	
)					GCC												3258
	Lys			Phe	Ala	Phe			Ile	Val	His	Leu	Tyr	Pro	Phe	Leu	
		100	0				100	5				1010	)				
10					GGT												3306
10			Leu	met	Gly			Asn	Arg	Thr			He	Val	Val		
	1015	•				102	J				1029	•				1030	
	TCC	<b>ም</b> ረጥ	OTT	CTC	TTG	COT	TH COTT	N TO	mmc.	***	mma	mmo	maa	- CMM	200	3 mm	2254
					Leu												3354
15	rrp	361	val	Deu	103		261	116	Pile	1040		Den	irp	vai	1049		
					105.	,				1041	•				104.	,	
	GAT	CCC	TTC	ACT	AGC	CGA	GTC	ACT	GGC	CCG	GAC	ATT	CTG	GAA	TGT	GGA	3402
					Ser												
	•			1050		•			1059		•			1060	_		
20																	
	ATC	AAC	TGT	TGAG	BAAG	CGA (	CAA	ATAT?	T AC	CTG	TTTY	AGC	GTT <i>I</i>	AAA			3451
	Ile	Asn	Cys														
			1065	5													
25	AAA	ACAC	AGA A	ATTT/	AAAT"	ra T	TTTT(	CATTO	G TT	TAT	ltgt	TCAC	TTT!	TT A	ACTT	TTGTTG	3511
	TGTC	TAT	CTG 7	rctg:	rtcg	rt C	rtcte	STCT	r gga	GTC	AATA	ATT	TATG	GT A	AGAA?	TATATC	3571
	TTAC	CTCT	AGT T	ract:	rtggi	AA AG	STTA?	TAAT	iaa 1	GTG	AAAG	CCA					3614
30																	
35	(2)	INF	)KMAT	LION	FOR	SEQ	to i	10:10	J:								
,,			(i) e	ייי	ENCE	CUAT	) N (~***	- D T C C	PT CC								
		,	11) 2	_	LEI						10						
				100	للظالد	·01U		, all		acit	437						

(B) TYPE: amino acid(D) TOPOLOGY: linear

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(xi)	SEQUENCE	DESCRIPTION:	SEO	ID	NO:10

5 Met Glu Ser Glu Gly Glu Thr Ala Gly Lys Pro Met Lys Asn Ile Val 1 5 10 15

Pro Gln Thr Cys Gln Ile Cys Ser Asp Asn Val Gly Lys Thr Val Asp
20 25 30

10

Gly Asp Arg Phe Val Ala Cys Asp Ile Cys Ser Phe Pro Val Cys Arg
35 40 45

Pro Cys Tyr Glu Tyr Glu Arg Lys Asp Gly Asn Gln Ser Cys Pro Gln 15 50 55 60

Cys Lys Thr Arg Tyr Lys Arg Leu Lys Gly Ser Pro Ala Ile Pro Gly 65 70 75 80

20 Asp Lys Asp Glu Asp Gly Leu Ala Asp Glu Gly Thr Val Glu Phe Asn
85 90 95

Tyr Pro Gln Lys Glu Lys Ile Ser Glu Arg Met Leu Gly Trp His Leu 100 105 110

25

Thr Arg Gly Lys Gly Glu Glu Met Gly Glu Pro Gln Tyr Asp Lys Glu 115 120 125

Val Ser His Asn His Leu Pro Arg Leu Thr Ser Arg Gln Asp Thr Ser 30 130 135 140

Gly Glu Phe Ser Ala Ala Ser Pro Glu Arg Leu Ser Val Ser Ser Thr 145 150 155 160

35 Ile Ala Gly Gly Lys Arg Leu Pro Tyr Ser Ser Asp Val Asn Gln Ser 165 170 175

Pro Asn Arg Arg Ile Val Asp Pro Val Gly Leu Gly Asn Val Ala Trp
180 185 190

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	Lys	Glu	Arg	Val	Asp	Gly	Trp	Lys	Met	Lys	Gln	Glu	Lys	Asn	Thr	Gly
			195					200					205			
	Pro	Val	Ser	Thr	Gln	Ala	Ala	Ser	Glu	Arg	Gly	Gly	Val	Asp	Ile	Asp
5		210					215					220				
	Ala	Ser	Thr	Asp	Ile	Leu	Ala	Asp	Glu	Ala	Leu	Leu	Asn	Asp	Glu	Ala
	225					230					235					240
10	Arg	Gln	Leu	Leu	Ser	Arg	Lys	Val	Ser	Ile	Pro	Ser	Ser	Arg	Ile	Asn
	_				245					250					255	
	Pro	Tyr	Arg	Met	Val	Ile	Met	Leu	Arg	Leu	Val	Ile	Leu	Cys	Leu	Phe
		•	_	260					265					270		
15																
	Leu	His	Tyr	Arg	Ile	Thr	Asn	Pro	Val	Pro	Asn	Ala	Phe	Ala	Leu	Trp
			275	_				280					285			
	Leu	Val	Ser	Val	Ile	Сув	Glu	Ile	Trp	Phe	Ala	Leu	Ser	Trp	Ile	Leu
20		290				•	295		•			300		-		
	Asp	Gln	Phe	Pro	Lvs	Trp	Phe	Pro	Val	Asn	Arg	Glu	Thr	Tyr	Leu	Asp
	305				•	310					315			•		320
25	Ara	Leu	Ala	Leu	Ara	Tvr	Asp	Arg	Glu	Glv	Glu	Pro	Ser	Gln	Leu	Ala
	3				325	-,-	•			330					335	
	Ala	Val	Asp	Ile	Phe	Val	Ser	Thr	Val	qsA	Pro	Leu	Lys	Glu	Pro	Pro
			•	340					345	•			•	350		
30																
	Leu	Val	Thr	Ala	Asn	Thr	Val	Leu	Ser	Ile	Leu	Ala	Val	Asp	Tyr	Pro
			355					360					365	•	•	
	Val	azA	Lvs	Val	Ser	Сув	Tyr	Val	Ser	qaA	qsA	Gly	Ala	Ala	Met	Leu
35		370	, -			•	375			•	•	380				
	Ser	Phe	Glu	Ser	Leu	Ala	Glu	Thr	Ser	Glu	Phe	Ala	Ara	Lvs	Tro	Val
	385					390					395		,	•	•	400

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	Pro	Phe	Cys	Lys	Lys 405	Tyr	Ser	Ile	Glu	Pro 410	Arg	Ala	Pro	Glu	Trp 415	Tyr
5	Phe	Ala	Ala	Lys 420	Ile	qaA	Tyr	Leu	Lys 425	Asp	Lys	Val	Gln	Thr 430	Ser	Phe
	Val	Lys	Asp 435	Arg	Arg	Ala	Met	Lys 440	Arg	Glu	Tyr	Glu	Glu 445	Phe	Lys	Ile
10	Arg	Ile 450	Asn	Ala	Leu	Val	Ser 455	Lys	Ala	Leu	Lys	Cys 460	Pro	Glu	Glu	Gly
15	Trp 465	Val	Met	Gln	Asp	Gly 470	Thr	Pro	Trp	Pro	Gly 475	Asn	Asn	Thr	Gly	Asp 480
	H18	Pro	Gly	Met	Ile 485	Gln	Val	Phe	Leu	Gly 490	Gln	Asn	Gly	Gly	Leu 495	Asp
20	Ala	Glu	Gly	Asn 500	Glu	Leu	Pro	Arg	Leu 505	Val	Tyr	Val	Ser	Arg 510	Glu	Lys
	Arg	Pro	Gly 515	Phe	Gln	His	His	Lys 520	Lys	Ala	Gly	Ala	Met 525	Asn	Ala	Leu
25	Val	Arg 530	Val	Ser	Ala	Val	Leu 535	Thr	Asn	Gly	Pro	Phe 540	Ile	Leu	Asn	Leu
30	Asp 545	Суз	Asp	His	Tyr	Ile 550	Asn	Asn	Ser	Lув	Ala 555	Leu	Arg	Glu	Ala	Met 560
	Сув	Phe	Leu	Met	Asp 565	Pro	Asn	Leu	Gly	Lys 570		Val	Сув	Tyr	Val 575	Gln
35	Phe	Pro	Gln	Arg 580	Phe	Aap	Gly	Ile	Asp 585	Lys	Asn	Asp	Arg	Tyr 590	Ala	Aan
	Arg	Asn	Thr 595	Val	Phe	Phe	Asp	Ile 600		Leu	Arg	Gly	Leu 605	Asp	Gly	Ile

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	Gln	Gly	Pro	Val	Tyr	Val	Gly	Thr	Gly	Сув	Val	Phe	Asn	Arg	Thr	Ala
		610					615					620				
	Leu	Tyr	Gly	Tyr	Glu	Pro	Pro	Ile	Lys	Val	Lys	His	Lys	Lys	Pro	Ser
5	625					630					635					640
	Leu	Leu	Ser	Lys	Leu	Cys	Gly	Gly	Ser	Arg	Lys	Lys	Asn	Ser	Lys	Ala
					645					650					655	
10	Lys	Lys	Glu	Ser	Asp	Lys	Lys	Lys	Ser	Gly	Arg	His	Thr	Asp	Ser	Thr
				660					665					670		
	Val	Pro	Val	Phe	Asn	Leu	Asp	Asp	Ile	Glu	Glu	Gly		Glu	Gly	Ala
			675					680					685			
15															_	
	Gly	Phe	Asp	qaA	Glu	Lys		Leu	Leu	Met	Ser		Met	Ser	Leu	Glu
		690					695					700				
						_		1	•	1		0	ml	•	Wa 5	G1
20		Arg	Phe	GIY	Gin		Ala	vaı	Pne	vaı	Ala	Ser	inr	Leu	Mec	720
20	705					710					715					720
	•	<b>0</b> 1	01		Due	Dun	C	210	Th.	Dvo	Glu	N a n	Dho	Lau	Lve	Glu
	ASN	GIY	GIY	vai		Pro	Ser	Ala	Int	730	GIU	Maii	FILE	neu	735	JIU
					725					,,,,						
25	Δla	Tle	Wi e	Val	11e	Ser	Cvs	Glv	Tvr	Glu	Asp	Lva	Ser	Asp	Trp	Glv
	ALG	110		740		501	-,-	<b></b> ,	745			-,-		750	•	•
	Met	Glu	Ile	Gly	Trp	Ile	Tyr	Gly	Ser	Val	Thr	Glu	Asp	Ile	Leu	Thr
			755	_	•		•	760					765			
30																
	Gly	Phe	Lys	Met	His	Ala	Arg	Gly	Trp	Arg	Ser	Ile	Tyr	Сув	Met	Pro
	-	770					775					780				
	Lys	Leu	Pro	Ala	Phe	Lys	Gly	Ser	Ala	Pro	Ile	Asn	Leu	Ser	Авр	Arg
35	785					790					795					800
	Leu	Asn	Gln	Val	Leu	Arg	Trp	Ala	Leu	Gly	Ser	Val	Glu	Ile	Leu	Phe
					805					810					815	

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	Ser	Arg	His	Cys 820	Pro	Ile	Trp	Tyr	Gly 825	Tyr	Asn	Gly	Arg	Leu 830	Lys	Phe
	Leu	Glu	Arg	Phe	Ala	Tyr	Val	Asn	Thr	Thr	Ile	Tyr	Pro	Ile	Thr	Ser
5			835					840					845			
	Ile	Pro 850	Leu	Leu	Met	Tyr	Сув 855	Thr	Leu	Leu	Ala	Val 860	Cys	Leu	Phe	Thr
10	Asn	Gln	Phe	Ile	Ile	Pro	Gln	Ile	Ser	Asn	Ile	Ala	Ser	Ile	Trp	Phe
	865					870					875					880
1.5	Leu	Ser	Leu	Phe	Leu 885	Ser	Ile	Phe	Ala	Thr 890	Gly	Ile	Leu	Glu	Met 895	Arg
15	Trp	Ser	Gly	Val 900	Gly	Ile	Asp	Glu	Trp 905	Тгр	Arg	Asn	Glu	Gln 910	Phe	Trp
20	Val	Ile	Gly 915	Gly	Val	Ser	Ala	His	Leu	Phe	Ala	Val	Phe 925	Gln	Gly	Ile
	Leu	Lys 930	Val	Leu	Ala	Gly	Ile 935	Asp	Thr	Asn	Phe	Thr 940	Val	Thr	Ser	Lys
25	Ala 945	Ser	Asp	Glu	qaA	Gly 950	Asp	Phe	Ala	Glu	Leu 955	Tyr	Leu	Phe	Lys	Trp 960
	Thr	Thr	Leu	Leu		Pro	Pro	Thr	Thr		Leu	Ile	Val	Asn		Val
30					965					970					975	
	Gly	Val	Val	Ala 980	Gly	Val	Ser	Tyr	Ala 985	Ile	Asn	Ser	Gly	Tyr 990	Gln	Ser
35	Trp	Gly	Pro 995		Phe	Gly	Lys	Leu 100		Phe	Ala	Phe	Trp		Ile	Val
			,,,						-				-00	-		
	His	Leu	_	Pro	Phe		Lys	-	Leu	Met	Gly	Arg		Asn	Arg	Thr

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1035

1040

60

Pro Thr Ile Val Val Val Trp Ser Val Leu Leu Ala Ser Ile Phe Ser

1030

1025

Leu Leu Trp Val Arg Ile Asp Pro Phe Thr Ser Arg Val Thr Gly Pro 5 1050 1045 Asp Ile Leu Glu Cys Gly Ile Asn Cys 1060 1065 10 (2) INFORMATION FOR SEQ ID NO:11: (i) SEQUENCE CHARACTERISTICS: 15 (A) LENGTH: 3673 base pairs (B) TYPE: nucleic acid (C) STRANDEDNESS: single (D) TOPOLOGY: linear 20 (ii) MOLECULE TYPE: cDNA (iii) HYPOTHETICAL: NO (iv) ANTI-SENSE: NO 25 (vi) ORIGINAL SOURCE: (A) ORGANISM: Arabidopsis thaliana (B) STRAIN: Columbia (C) INDIVIDUAL ISOLATE: rswl mutant 30 (ix) FEATURE: (A) NAME/KEY: CDS (B) LOCATION: 71..3313 35 (xi) SEQUENCE DESCRIPTION: SEQ ID NO:11:

GAATCGGCTA CGAATTTCCC AATTTTGAAT TTTGTGAATC TCTCTCTTTC TCTGTGTGTC

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	GGTG	GCT	GCG .	ATG	GAG	GCC	AGT	GCC	GGC '	TTG	GTT	GCT	GGA	TCC	TAC	CGG	109
				Met	Glu	Ala	Ser	Ala	Gly	Leu	Val	Ala	Gly	Ser	Tyr	Arg	
				1				5					10				
5	ACA.	244	GAG	CTC	י כדיד	. GCG	ב אירר	CCA	ר מת	GAA	TCT	י האיז	r eec	ccc	ACC	מממ	157
_															Thr		15,
	9	15	014	200			20	_		014	JCI	25	_	U.,		Буб	
	CCT	TTG	AAG	CAA	TATG	TAA 3	GGC	CAG	ATA	TGT	CAG	ATO	TGT	GGI	GAT	GAT	205
10	Pro	Leu	Lys	Asr	Met	Asr	Gly	Gln	Ile	Суз	Gln	Ile	cys	Gly	' Asp	Asp	
	30					35	•				40	)				45	
	CTT	GGA	CTC	הנים	<b>445)</b>	אריז	· GGA	СУТ	. GTC	andra.	· GTC		י ייניי	דממ י	GAA	тст	253
															Glu		233
15					50		•			55			•		60	•	
	GCC	TTC	CCT	GTO	TGT	. CGG	CCT	TGC	TAT	GAG	TAC	GAC	AGG	AAA :	GAT	GGA	301
	Ala	Phe	Pro	Va]	l Сув	Arg	Pro	Сув	Tyr	Glu	Туг	Glu	a Arg	Lys	Asp	Gly	
20				65	5				70					75	5		
20	ΔСТ	CAG	ጥርጥ	TGO	י רכייו	י ראז	י דכר	. אאר	ייט א	) (C)	ጥጥር	י אכי	CG	CDC	י אככ	GGG	349
																Gly	343
			80	-		-	•	85		-			90			,	
25	AGT	ССТ	CGT	GT	r gaa	GG	A GAT	GAA	GAT	GAG	GAT	GA!	r GTI	GAT	GAT	ATC	397
	Ser	Pro	Arg	Va]	l Glu	Gly	/ Asp	Glu	Asp	Glu	Asp	As <sub>I</sub>	Val	. Asr	Asp	Ile	
		95					100					109	5				
	GAG	דממ	GAG	. TT(	רממ ־	מד י	י מכר	' CAG	GGA	GCT	י אמר	יממי	3 GCC	י אכי	י ראר	CAA	445
30																Gln	113
	110					119			•		120			•		125	
	CGC	CAT	GGC	GA.	A GAG	TT	r TCT	TC1	TCC	TCI	AGA	CA'	r gaa	TC	CAA	CCA	493
	Arg	His	Gly	Gli	u Glu	ı Phe	e Ser	Ser	Ser	Ser	Arg	, Hi	s Glu	sez	r Glm	Pro	
35					130	)				135	5				140	)	
	<u>አ</u> ጥጥ	درسد	سنس	، سب ا	ר ארי	י ראי	ר כככ	י ראי	ר ארר	GT <sup>n</sup>	ր գր⁄աս	ר ממי	a (12)	2 2 1000	r	ACG	E 4 1
																Thr	541
				14			,		150				,	159	_	• • •	
40																	

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	CCT	GAT	ACA	CAA	TCT	GTG	CGA	ACT	ACA	TCA	GGT	CCT	TTG	GGT	CCT	TCT	589
	Pro	Asp	Thr	Gln	Ser	Val	Arg	Thr	Thr	Ser	Gly	Pro	Leu	Gly	Pro	Ser	
			160					165					170				
5	GAC	AGG	AAT	GCT	ATT	TCA	TCT	CCA	TAT	ATT	GAT	CCA	CGG	CAA	CCT	GTC	637
	Asp	Arg	Asn	Ala	Ile	Ser	Ser	Pro	Tyr	Ile	Asp	Pro	Arg	Gln	Pro	Val	
		175					180					185					
	CCT	GTA	AGA	ATC	GTG	GAC	CCG	TCA	AAA	GAC	TTG	AAC	TCT	TAT	GGG	CTT	685
10	Pro	Val	Arg	Ile	Val	Asp	Pro	Ser	Lys	Asp	Leu	Asn	Ser	Tyr	Gly	Leu	
	190					195					200					205	
	GGT	AAT	GTT	GAC	TGG	AAA	GAA	AGA	GTT	GAA	GGC	TGG	AAG	CTG	AAG	CAG	733
	Gly	Asn	Val	Asp	Trp	Lys	Glu	Arg	Val	Glu	Gly	Trp	Lys	Leu	Lys	Gln	
15					210					215					220		
	GAG	AAA	AAT	ATG	TTA	CAG	ATG	ACT	GGT	AAA	TAC	CAT	GAA	GGG	AAA	GGA	781
	Glu	Lys	Asn	Met	Leu	Gln	Met	Thr	Gly	Lув	Tyr	His	Glu	Gly	Lys	Gly	
				225					230					235			
20																	
	GGA	GAA	ATT	GAA	GGG	ACT	GGT	TCC	AAT	GGC	GAA	GAA	CTC	CAA	ATG	GCT	829
	Gly	Glu	Ile	Glu	Gly	Thr	Gly	Ser	Asn	Gly	Glu	Glu	Leu	Gln	Met	Ala	
			240					245					250				
25	GAT	GAT	ACA	CGT	CTT	ССТ	ATG	AGT	CGT	GTG	GTG	CCT	ATC	CCA	TCT	TCT	877
	Asp	Asp	Thr	Arg	Leu	Pro	Met	Ser	Arg	Val	Val	Pro	Ile	Pro	Ser	Ser	
		255					260					265					
	CGC	CTA	ACC	ССТ	TAT	CGG	GTT	GTG	ATT	ATT	CTC	CGG	CTT	ATC	ATC	TTG	925
30	Arg	Leu	Thr	Pro	Tyr	Arg	Val	Val	Ile	Ile	Leu	Arg	Leu	Ile	Ile	Leu	
	270					275					280					285	
	TGT	TTC	TTC	TTG	CAA	TAT	CGT	ACA	ACT	CAC	ССТ	GTG	AAA	AAT	GCA	TAT	973
	Cys	Phe	Phe	Leu	Gln	Tyr	Arg	Thr	Thr	His	Pro	Val	Lys	Asn	Ala	Tyr	
35					290					295					300		
	ССТ	TTG	TGG	TTG	ACC	TCG	GTT	ATC	TGT	GAG	ATC	TGG	TTT	GCA	TTT	TCT	1021
	Pro	Leu	Trp	Leu	Thr	Ser	Val	Ile	Сув	Glu	Ile	Trp	Phe	Ala	Phe	Ser	
			-	305					310			-		315			

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	TGG	CTT	CTT	GAT	CAG	TTT	CCC	AAA	TGG	TAC	CCC	ATT	AAC	AGG	GAG	ACT	1069
	Trp	Leu	Leu	qaA	Gln	Phe	Pro	Lys	Trp	Tyr	Pro	Ile	Asn	Arg	Glu	Thr	
			320					325					330				
5	TAT	CTT	GAC	CGT	CTC	GCT	ATA	AGA	TAT	GAT	CGA	GAC	GGT	GAA	CCA	TCA	1117
	Tyr	Leu	Asp	Arg	Leu	Ala	Ile	Arg	Tyr	Asp	Arg	Asp	Gly	Glu	Pro	Ser	
		335					340					345					
	CAG	CTC	GTT	CCT	GTT	GAT	GTG	TTT	GTT	AGT	ACA	GTG	GAC	CCA	TTG	AAA	1165
10	Gln	Leu	Val	Pro	Val	Asp	Val	Phe	Val	Ser	Thr	Val	Asp	Pro	Leu	Lys	
	350					355					360					365	
	GAG	CCT	CCC	CTT	GTT	ACA	GCA	AAC	ACA	GTT	CTC	TCG	ATT	CTT	TCT	GTG	1213
	Glu	Pro	Pro	Leu	Val	Thr	Ala	Asn	Thr	Val	Leu	Ser	Ile	Leu	Ser	Val	
15					370					375					380		
	GAC	TAC	CCG	GTA	GAT	AAA	GTA	GCC	TGT	TAT	GTT	TCA	GAT	GAT	GGT	TCA	1261
	Asp	Tyr	Pro	Val	Asp	ГÀа	Val	Ala	Cys	Tyr	Val	Ser	Asp	Asp	Gly	Ser	
				385					390					395			
20																	
	GCT	ATG	CTT	ACC	TTT	GAA	TCC	CTT	TCT	GAA	ACC	GCT	GAG	TTT	GCA	AAG	1309
	Ala	Met	Leu	Thr	Phe	Glu	Ser	Leu	Ser	Glu	Thr	Ala	Glu	Phe	Ala	Lys	
			400					405					410				
25																CCT	1357
	Lys	Trp	Val	Pro	Phe	Cys	Lys	Lys	Phe	Asn	Ile	Glu	Pro	Arg	Ala	Pro	
		415					420					425					
	GAA	TTC	TAT	TTT	GCC	CAG	AAG	ATA	GAT	TAC	TTG	AAG	GAC	AAG	ATC	CAA	1405
30	Glu	Phe	Tyr	Phe	Ala	Gln	Lys	Ile	Asp	Tyr	Leu	Lys	qaA	Lys	Ile	Gln	
	430					435					440	ı				445	
	CCG	TCT	TTT	GTT	AAA	GAG	CGA	CGA	GCT	ATG	AAG	AGA	GAG	TAT	GAA	GAG	1453
		Ser	Phe	Val	Lys	Glu	Arg	Arg	Ala	Met	Lys	Arg	Glu	Туг	Glu	Glu	
35					450					455					460	ı	
	TTT	AAA	GTG	AGG	ATA	CAA .	GCI	CTI	GTT	GCC	. AAA	GCA	CAG	AA.	ATC	CCT	1501
	Phe	Lys	val	. Arg	Ile	Asn	Ala	Lev	Val	Ala	Lys	a Ala	Glr	Lys	ı Ile	Pro	
				465	<b>i</b>				470	ı				475	5		
40	1																

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GAA	GAA	GGC	TGG	ACA	ATG	CAG	GAT	GGT	ACT	CCC	TGG	CCT	GGT	AAC	AAC	1549
Glu	Glu	Gly	Trp	Thr	Met	Gln	Asp	Gly	Thr	Pro	Trp	Pro	Gly	Asn	Asn	
		480					485					490				
ACT	AGA	GAT	CAT	CCT	GGA	ATG	ATA	CAG	GTG	TTC	TTA	GGC	CAT	AGT	GGG	1597
Thr	Arg	Asp	His	Pro	Gly	Met	Ile	Gln	Val	Phe	Leu	Gly	His	Ser	Gly	
	495					500					505					
GGT	CTG	GAT	ACC	GAT	GGA	AAT	GAG	CTG	CCT	AGA	CTC	ATC	TAT	GTT	TCT	1645
Gly	Leu	Asp	Thr	Asp	Gly	Asn	Glu	Leu	Pro	Arg	Leu	Ile	Tyr	Val	Ser	
510					515					520					525	
CGT	GAA	AAG	CGG	CCT	GGA	TTT	CAA	CAC	CAC	AAA	AAG	GCT	GGA	GCT	ATG	1693
Arg	Glu	Lys	Arg	Pro	Gly	Phe	Gln	His	His	Lys	Lys	Ala	Gly	Ala	Met	
				530					535					540		
AAT	GCA	TTG	ATC	CGT	GTA	TCT	GTT	GTT	CTT	ACC	AAT	GGA	GCA	TAT	CTT	1741
Asn	Ala	Leu	Ile	Arg	Val	Ser	Val	Val	Leu	Thr	Asn	Gly	Ala	Tyr	Leu	
			545					550					555			
TTG	AAC	GTG	GAT	TGT	GAT	CAT	TAC	TTT	AAT	AAC	AGT	AAG	GCT	ATT	AAA	1789
Leu	Asn	Val	Asp	Cys	Asp	His	Tyr	Phe	Asn	Asn	Ser	Lys	Ala	Ile	Lys	
		560					565					570				
GAA	GCT	ATG	TGT	TTC	ATG	ATG	GAC	CCG	GCT	ATT	GGA	AAG	AAG	TGC	TGC	1837
Glu	Ala	Met	Cys	Phe	Met	Met	Asp	Pro	Ala	Ile	Gly	Lys	Lys	Сув	Cys	
	575					580					585					
TAT	GTC	CAG	TTC	CCT	CAA	CGT	TTT	GAC	GGT	ATT	GAT	TTG	CAC	GAT	CGA	1885
Tyr	Val	Gln	Phe	Pro	Gln	Arg	Phe	Asp	Gly	Ile	Asp	Leu	His	Asp	Arg	
590					595					600					605	
TAT	GCC	AAC	AGG	AAT	ATA	GTC	TTT	TTC	GAT	ATT	AAC	ATG	AAG	GGG	TTG	1933
Tyr	Ala	Asn	Arg	Asn	Ile	Val	Phe	Phe	Asp	Ile	Asn	Met	Lys	Gly	Leu	
				610					615					620		
GAT	GGT	ATC	CAG	GGT	CCA	GTA	TAT	GTG	GGT	ACT	GGT	TGT	TGT	TTT	AAT	1981
Asp	Gly	Ile	Gln	Gly	Pro	Val	Tyr	Val	Gly	Thr	Gly	Cys	Сув	Phe	Asn	
			625					630					635			
	Glu ACT Thr GGT Gly 510 CGT Arg AAT Asn TTG Leu GAA Glu TAT Tyr 590 TAT Tyr	Glu Glu  ACT AGA Thr Arg 495  GGT CTG Gly Leu 510  CGT GAA Arg Glu  AAT GCA Asn Ala  TTG AAC Leu Asn  GAA GCT Glu Ala 575  TAT GTC Tyr Val 590  TAT GCC Tyr Ala	Glu Glu Gly 480  ACT AGA GAT Thr Arg Asp 495  GGT CTG GAT Gly Leu Asp 510  CGT GAA AAG Arg Glu Lys  AAT GCA TTG Asn Ala Leu  TTG AAC GTG Leu Asn Val 560  GAA GCT ATG Glu Ala Met 575  TAT GTC CAG Tyr Val Gln 590  TAT GCC AAC Tyr Ala Asn	Glu Glu Gly Trp 480  ACT AGA GAT CAT Thr Arg Asp His 495  GGT CTG GAT ACC Gly Leu Asp Thr 510  CGT GAA AAG CGG Arg Glu Lys Arg  AAT GCA TTG ATC Asn Ala Leu Ile 545  TTG AAC GTG GAT Leu Asn Val Asp 560  GAA GCT ATG TGT Glu Ala Met Cys 575  TAT GTC CAG TTC Tyr Val Gln Phe 590  TAT GCC AAC AGG Tyr Ala Asn Arg  GAT GGT ATC CAG Asp Gly Ile Gln	Glu Glu Gly Trp Thr 480  ACT AGA GAT CAT CCT Thr Arg Asp His Pro 495  GGT CTG GAT ACC GAT Gly Leu Asp Thr Asp 510  CGT GAA AAG CGG CCT Arg Glu Lys Arg Pro 530  AAT GCA TTG ATC CGT Asn Ala Leu Ile Arg 545  TTG AAC GTG GAT TGT Leu Asn Val Asp Cys 560  GAA GCT ATG TGT TTC Glu Ala Met Cys Phe 575  TAT GTC CAG TTC CCT Tyr Val Gln Phe Pro 590  TAT GCC AAC AGG AAT Tyr Ala Asn Arg Asn 610  GAT GGT ATC CAG GGT Asp Gly Ile Gln Gly	Glu Glu Gly Trp Thr Met	Glu         Glu         Gly         Trp         Thr         Met         Gln           ACT         AGA         GAT         CCT         GGA         ATG           Thr         Arg         Asp         His         Pro         Gly         Met           495         ACC         GAT         GGA         AAT           GGT         CTG         GAT         ACC         GAT         GGA         AAT           G1y         Leu         Asp         Thr         Asp         Gly         Asn           S10         Leu         Asp         CCT         GGA         TTT           AAT         GLu         Lys         Arg         Pro         Gly         Phe           AAT         ASA         CGG         CCT         GAT         CCT           AAT         ASA         ASA         ASA         CGT         GTA         CAT           AAT         ASA         ASA         CGT         GTA         CAT         CAT           AAT         ASA         ASA         ASA         CAT         ATG         ATG         ATG         ATG         ATG         ATG         ATG         ATG         ATG         ATG	Glu Glu Gly Trp Thr Met Gln Asp	Glu Glu Gly 480         Trp Thr Met Gln Asp Gly 485           ACT AGA GAT CAT CCT GGA ATG ATA CAG Thr Arg Asp His Pro Gly Met 495         Gly Met Ile Gln 500           GGT CTG GAT ACC GAT GGA AAT GAG CTG Gly Leu Asp Thr Asp Gly Asn Glu Leu 510         GGY ARG GRA AAG CGG CCT GGA TTT CAA CAC ARG Glu Lys Arg Pro Gly Phe Gln His 530           AAT GCA TTG ATC CGT GTA TCT GTT Asn Ala Leu Ile Arg Val Ser Val Val 545         550           TTG AAC GTG GAT TGT GAT CAT TAC TTT Leu Asn Val Asp Cys Asp His Tyr Phe 565         TTG AGC ATG TGT TTC ATG ATG GAC CCG Glu Ala Met Cys Phe Met Met Asp Pro 575           GAA GCT ATG TGT CCT CAA CGT TTT GAC Tyr Val Gln Phe Pro Gln Arg Phe Asp 590         TTC CAG AAT ATA GTC TTT TTC ATG ATG ATG Phe Asp 590           TAT GCC AAC AGG AAT ATA GTC TTT TTC ATG ATG ATG GAC CCG GATA TAT GTC TYR Ala Asn Arg Asn Ile Val Phe Phe 610           GAT GGT ATC CAG GGT CCA GTA TAT GTG ASp Gly Ile Gln Gly Pro Val Tyr Val	Glu         Glu         Gly         Trp         Thr         Met         Gln         Asp         Gly         Thr           ACT         AGA         GAT         CCT         GGA         ATG         ATA         CAG         GTG           Thr         Arg         Asp         His         Pro         Gly         Met         Ile         GIn         Val           GGT         CTG         GAT         ACC         GAT         GGA         AAT         GAG         CTG         CCT           GJY         Leu         Asp         Thr         Asp         GJY         Asn         GJU         Leu         Pro         GJU         Asn         CAC         CAC         CAC         AAT         CAC         CAC	Glu         Glu         Gly         Trp         Thr         Met         Gln         Asp         Gly         Thr         Pro           ACT         AGA         GAT         CAT         CCT         GGA         ATA         CAG         GTG         TTC           Thr         Arg         Asp         His         Pro         Gly         Met         Ile         Gln         Val         Phe           GGT         CTG         GAT         ACC         GAT         GGA         AAT         GAG         CTG         AGA           G1y         Leu         Asp         Thr         Asp         Gly         Asn         Glu         Leu         Pro         Arg           510         Leu         Asp         Thr         Asp         Gly         Asn         Glu         Leu         Pro         Arg           CGT         GAA         AAG         CGG         CCT         GGA         TTT         CAA         CAC         CAC         AAA           AAT         GCA         ATG         ATG         CTT         CTT         CTT         ACC         CAC         AAA         AAC         AAC         AAC         AAC         AAC	Glu Glu Gly Trp Thr Met Gln Asp Gly Thr Pro Trp	Glu Glu Gly Trp Thr Met Gln Asp Gly Thr Pro Trp Pro 480	Glu Glu Gly Trp Thr Met Gln Asp Gly Thr Pro Trp Pro Gly 480	Glu Glu Gly Trp Thr Met Gln Asp Gly Thr Pro Trp Pro Gly Asn 480	ACT AGA GAT CAT CCT GGA ATG ATA CAG GTG TTC TTA GGC CAT AGT GGG Thr Arg Asp His Pro Gly Met 11e Gln Val Phe Leu Gly His Ser Gly 495 500 505  GGT CTG GAT ACC GAT GGA AAT GAG CTG CCT AGA CTC ATC TAT GTT TCT Gly Leu Asp Thr Asp Gly Asn Glu Leu Pro Arg Leu 11e Tyr Val Ser 510 515 520 525  CGT GAA AAG CGG CCT GGA TTT CAA CAC CAC AAA AAG GCT GGA GCT ATG Arg Glu Lys Arg Pro Gly Phe Gln His His Lys Lys Ala Gly Ala Met 530 535 540  AAT GCA TTG ATC CGT GTA TCT GTT GTT CTT ACC AAT GGA GCA TAT CTT Asn Ala Leu 11e Arg Val Ser Val Val Leu Thr Asn Gly Ala Tyr Leu 545 550 555  TTG AAC GTG GAT TGT GAT CAT TAC TTT AAT AAC AGT AAG GCT ATT AAA Leu Asn Val Asp Cys Asp His Tyr Phe Asn Asn Ser Lys Ala 11e Lys 560 565 570  GAA GCT ATG TGT TTC ATG ATG GAC CCG GCT ATT GGA AAG AAG TGC TGC Glu Ala Met Cys Phe Met Met Asp Pro Ala 11e Gly Lys Lys Cys Cys 575 580 585  TAT GTC CAG TTC CCT CAA CGT TTT GAC GGT ATT GAT TG CAC GAT CGA Tyr Val Gln Phe Pro Gln Arg Phe Asp Gly Ile Asp Leu His Asp Arg 590 595 600  GAT GCC AAC AGG AAT ATA GTC TTT TTC GAT ATT AAC ATG AAG GGG TTG Tyr Ala Asn Arg Asn Ile Val Phe Phe Asp Ile Asn Met Lys Gly Leu 610 615 620

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	AGG	CAG	GCT	CTA	TAT	GGG	TAT	GAT	CCT	GTT	TTG	ACG	GAA	GAA	GAT	ATT	2029
	Arg	Gln	Ala	Leu	Tyr	Gly	Tyr	Asp	Pro	Val	Leu	Thr	Glu	Glu	Asp	Leu	
			640					645					650				
_																	
5	GAA	CCA	TAA	ATT	ATT	GTC	AAG	AGC	TGT	TGC	GGG	TCA	AGG	AAG	AAA	GGT	2077
	Glu	Pro	Asn	Ile	Ile	Val	Lys	Ser	Cys	Cys	Gly	Ser	Arg	Lув	Lys	Gly	
		655					660					665					
10				AAG													2125
10		Ser	Ser	Lys	Lys		Asn	Tyr	Glu	Lys		Arg	Gly	Ile	Asn		
	670					675					680					685	
	አረተ	CNC	TOO	n n m	com	CCN	<b>O</b> TTO	mmc.	2.20	N TOC	020	<b>a.</b> a	200	C. M.	~~~		
				AAT													2173
15	361	wah	261	Asn	690	PLO	Den	PHE	ASII		GIU	wab	116	мар		GIY	
13					090					695					700		
	TTT	GAA	GGT	TAT	GAT	GAT	GAG	AGG	тст	ATT	СТА	ATG	TCC	CAG	AGG	AGT	2221
				Tyr													
				705		•			710					715			
20																	
	GTA	GAG	AAG	CGT	TTT	GGT	CAG	TCG	CCG	GTA	TTT	ATT	GCG	GCA	ACC	TTC	2269
	Val	Glu	Lys	Arg	Phe	Gly	Gln	Ser	Pro	Val	Phe	Ile	Ala	Ala	Thr	Phe	
			720					725					730				
25	ATG	GAA	CAA	GGC	GGC	ATT	CCA	CCA	ACA	ACC	AAT	CCC	GCT	ACT	CTT	CTG	2317
	Met	Glu	Gln	Gly	Gly	Ile	Pro	Pro	Thr	Thr	Asn	Pro	Ala	Thr	Leu	Leu	
		735					740					745					
••				ATT													2365
30	Lys	Glu	Ala	Ile	His	Val	Ile	Ser	Сув	Gly	Tyr	Glu	Asp	Lys	Thr	Glu	
	750					755					760					765	
				GAG													2413
25	Trp	Gly	Lys	Glu		Gly	Trp	Ile	Tyr		Ser	Val	Thr	Glu	-	Ile	
35					770					775					780		
	Curur	አ <i>ር</i> ም	ccc	doub.	ת ת	איזירי	(13 Tr	CCC	CCC	CCm	mc c	n m r	TCC.	200	ma c	TOC	2451
				TTC									-				2461
	ມຮູດ	THE	GIA	Phe	ьyв	met	nis	WIG		GIA	ırp	тте	ser		Tyr	сув	
				785					790					795			

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	AAT	CCT	CCA	CGC	сст	GCG	TTC	AAG	GGA	TCT	GCA	CCA	ATC	AAT	CTT	TCT	2509
	Asn	Pro	Pro	Arg	Pro	Ala	Phe	Lys	Gly	Ser	Ala	Pro	Ile	Asn	Leu	Ser	
			800					805					810				
5	GAT	CGT	TTG	AAC	CAA	GTT	CTT	CGA	TGG	GCT	TTG	GGA	TCT	ATC	GAG	ATT	2557
	Asp	Arg	Leu	Asn	Gln	Val	Leu	Arg	Trp	Ala	Leu	Gly	Ser	Ile	Glu	Ile	
		815					820					825					
	CTT	CTT	AGC	AGA	CAT	TGT	CCT	ATC	TGG	TAT	GGT	TAC	CAT	GGA	AGG	TTG	2605
10	Leu	Leu	Ser	Arg	His	Cys	Pro	Ile	Trp	Tyr	Gly	Tyr	His	Gly	Arg	Leu	
	830					835					840					845	
	AGA	CTT	TTG	GAG	AGG	ATC	GCT	TAT	ATC	AAC	ACC	ATC	GTC	TAT	CCT	ATT	2653
	Arg	Leu	Leu	Glu	Arg	Ile	Ala	Tyr	Ile	Asn	Thr	Ile	Val	Tyr	Pro	Ile	
15					850					855					860		
	ACA	TCC	ATC	CCT	CTT	ATT	GCG	TAT	TGT	ATT	CTT	CCC	GCT	TTT	TGT	CTC	2701
	Thr	Ser	Ile	Pro	Leu	Ile	Ala	Tyr	Сув	Ile	Leu	Pro	Ala	Phe	Cys	Leu	
				865					870					875			
20																	
	ATC	ACC	GAC	AGA	TTC	ATC	ATA	ccc	GAG	ATA	AGC	AAC	TAC	GCG	AGT	ATT	2749
	Ile	Thr	Asp	Arg	Phe	Ile	Ile	Pro	Glu	Ile	Ser	Asn	Tyr	Ala	Ser	Ile	
			880					885					890				
25	TGG	TTC	ATT	CTA	CTC	TTC	ATC	TCA	ATT	GCT	GTG	ACT	GGA	ATC	CTG	GAG	2797
	Trp	Phe	Ile	Leu	Leu	Phe	Ile	Ser	Ile	Ala	Val	Thr	Gly	Ile	Leu	Glu	
		895					900					905					
	CTG	AGA	TGG	AGC	GGT	GTG	AGC	ATT	GAG	GAT	TGG	TGG	AGG	AAC	GAG	CAG	2845
30	Leu	Arg	Trp	Ser	Gly	Val	Ser	Ile	Glu	qaA	Trp	Trp	Arg	Asn	Glu	Gln	
	910					915					920					925	
	TTC	TGG	GTC	ATT	GGT	GGC	ACA	TCC	GCC	CAT	CTT	TTT	GCT	GTC	TTC	CAA	2893
	Phe	Trp	Val	Ile	Gly	Gly	Thr	Ser	Ala	His	Leu	Phe	Ala	Val	Phe	Gln	
35					930					935					940		
	GGT	CTA	CTT	AAG	GTT	CTT	GCT	GGT	ATC	GAC	ACC	AAC	TTC	ACC	GTT	ACA	2941
	Gly	Leu	Leu	Lys	Val	Leu	Ala	Gly	Ile	qaA	Thr	Asn	Phe	Thr	Val	Thr	
				945					950					955			

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	TCT	AAA	GCC	ACA	GAC	GAA	GAT	GGG	GAT	TTT	GCA	GAA	CTC	TAC	ATC	TTC	2989
	Ser	Lys	Ala	Thr	Asp	Glu	Asp	Gly	Asp	Phe	Ala	Glu	Leu	Tyr	Ile	Phe	
			960					965					970				
5	AAA	TGG	ACA	GCT	CTT	CTC	ATT	CCA	CCA	ACC	ACC	GTC	CTA	CTT	GTG	AAC	3037
	Lys	Trp	Thr	Ala	Leu	Leu	Ile	Pro	Pro	Thr	Thr	Val	Leu	Leu	Val	Asn	
		975					980					985					
	CTC	ATA	GGC	ATT	GTG	GCT	GGT	GTC	TCT	TAT	GCT	GTA	AAC	AGT	GGC	TAC	3085
10	Leu	Ile	Gly	Ile	Val	Ala	Gly	Val	Ser	Tyr	Ala	Val	Asn	Ser	Gly	Tyr	
	990					995					1000	0				1005	
	CAG	TCG	TGG	GGT	CCG	CTT	TTC	GGG	AAG	CTC	TTC	TTC	GCC	TTA	TGG	GTT	3133
	Gln	Ser	Trp	Gly	Pro	Leu	Phe	Gly	Lys	Leu	Phe	Phe	Ala	Leu	Trp	Val	
15					1010	ס				1019	5				102	0	
	ATT	GCC	CAT	CTC	TAC	CCT	TTC	TTG	AAA	GGT	CTG	TTG	GGA	AGA	CAA	AAC	3181
	Ile	Ala	His	Leu	Tyr	Pro	Phe	Leu	Lys	Gly	Leu	Leu	Gly	Arg	Gln	Asn	
				102	5				1030	)				103	5		
20																	
	CGA	AÇA	CCA	ACC	ATC	GTC	ATT	GTC	TGG	TCT	GTT	CTT	CTC	GCC	TCC	ATC	3229
	Arg	Thr	Pro	Thr	Ile	Val	Ile	Val	Trp	Ser	Val	Leu	Leu	Ala	Ser	Ile	
			1040	0				104	5				1056	D			
25	TTC	TCG	TTG	CTT	TGG	GTC	AGG	ATC	AAT	CCC	TTT	GTG	GAC	GCC	AAT	ccc	3277
					Trp												
		105	5		•		1060	0				1069	5				
	AAT	GCC	AAC	AAC	TTC	AAT	GGC	AAA	GGA	GGT	GTC	TTT	TAG	ACCC	TAT		3323
30					Phe												
	107					107		•	•	•	108						
	TTA	IATA	CTT (	GTGT	GTGC	AT A'	TATC	AAAA	A CG	CGCA	ATGG	GAA'	TTCC	AAA '	TCAT	CTAAAC	3383
												_					
35	CCA'	TCAA	ACC (	CCAG	TGAA	CC G	GGCA	GTTA	A GG	rgat'	TCCA	TGT	CCAA	GAT '	TAGC	тттстс	3443
	CGA	GTAG	CCA (	GAGA.	AGGT	GA A	ATTG:	TTCG'	T AA	CACT	ATTG	TAA	TGAT"	TTT	CCAG'	TGGGGA	3503
			_ •				_ 3.	20									
	AGA	AGAT	GTG (	GACC	CAAA'	TG A'	TACA'	TAGT	C TA	CAAA	AAGA	ATT	TGTT	ATT	CTTT	CTTATA	3563
40			-											-			
-																	

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TTTATTTTAT TTAAAGCTTG TTAGACTCAC ACTTATGTAA TGTTGGAACT TGTTGTCCTA 3623 AAAAGGGATT GGAGTTTTCT TTTTATCTAA GAATCTGAAG TTTATATGCT 3673 5 (2) INFORMATION FOR SEQ ID NO:12: 10 (i) SEQUENCE CHARACTERISTICS: (A) LENGTH: 1081 amino acids (B) TYPE: amino acid (D) TOPOLOGY: linear 15 (11) MOLECULE TYPE: protein (xi) SEQUENCE DESCRIPTION: SEQ ID NO:12: Met Glu Ala Ser Ala Gly Leu Val Ala Gly Ser Tyr Arg Arg Asn Glu 20 ı 10 15 Leu Val Arg Ile Arg His Glu Ser Asp Gly Gly Thr Lys Pro Leu Lys 25 30 20 25 Asn Met Asn Gly Gln Ile Cys Gln Ile Cys Gly Asp Asp Val Gly Leu Ala Glu Thr Gly Asp Val Phe Val Ala Cys Asn Glu Cys Ala Phe Pro 55 30 Val Cys Arg Pro Cys Tyr Glu Tyr Glu Arg Lys Asp Gly Thr Gln Cys 70 75 Cys Pro Gln Cys Lys Thr Arg Phe Arg Arg His Arg Gly Ser Pro Arg 35 85 90 Val Glu Gly Asp Glu Asp Glu Asp Asp Val Asp Asp Ile Glu Asn Glu

105

110

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	Phe	Asn	Tyr	Ala	Gln	Gly	Ala	Asn	Lys	Ala	Arg	His	Gln	Arg	His	Gly
			115					120					125			
	Glu	Glu	Phe	Ser	Ser	Ser	Ser	Arg	His	Glu	Ser	Gln	Pro	Ile	Pro	Leu
5		130					135					140				
	Leu	Thr	His	Gly	His	Thr	Val	Ser	Gly	Glu	Ile	Arg	Thr	Pro	Asp	Thr
	145					150					155					160
10	Gln	Ser	Va 1	Ara	Thr	Thr	Ser	Glv	Pro	I.eu	Glv	Pro	Sar	Acn	D. T.C.	Aan
••	<b>G1</b>	261	<b>V</b>	n. g	165	1111	361	GIY	110	170	Gly	FEU	361	vob	175	Aon
	Ala	Ile	Ser	Ser	Pro	Tyr	Ile	Asp	Pro	Arg	Gln	Pro	Val	Pro	Val	Arg
				180					185					190		
15			_	_	_	_	_	_	_	_	_		_			
	He	Vai	195	Pro	Ser	Lys	Авр	200	Asn	ser	Tyr	Gly	Leu 205	Gly	Asn	Val
			1,,,					200					203			
	Asp	Trp	Lys	Glu	Arg	Val	Glu	Gly	Trp	Lys	Leu	Lys	Gln	Glu	Lys	Asn
20		210					215					220				
		Leu	Gln	Met	Thr	Gly	Lys	Tyr	His	Glu	_	Lys	Gly	Gly	Glu	
	225					230					235					240
25	Glu	Gly	Thr	Gly	Ser	Asn	Gly	Glu	Glu	Leu	Gln	Met	Ala	Asp	Авр	Thr
					245					250					255	
	Arg	Leu	Pro		Ser	Arg	Val	Val		Ile	Pro	Ser	Ser	•	Leu	Thr
30				260					265					270		
50	Pro	Tyr	Arg	Val	Val	Ile	Ile	Leu	Arq	Leu	Ile	Ile	Leu	Cvs	Phe	Phe
		•	275					280					285			
	Leu	Gln	Tyr	Arg	Thr	Thr	His	Pro	Val	Lys	Asn	Ala	Tyr	Pro	Leu	Trp
35		290					295					300				
	I.A.	ጥኩ 🖛	Qn=	Va.	71.	C	G3 v	71.	Terr	Dhe	<b>71</b> -	pha	C	Т	1	¥ =::
	305		Set	val	116	Сув 310		116	тър	Fne	315		ser	ırp	rea	Leu 320

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	Asp	Gln	Phe	Pro	Lys	Trp	Tyr	Pro	Ile	Asn	Arg	Glu	Thr	Tyr	Leu	Asp
					325					330					335	
	Arg	Leu	Ala	Ile	Arg	Tyr	Asp	Arg	Asp	Gly	Glu	Pro	Ser	Gln	Leu	Val
5	_			340	_	-	-	_	345					350		
-																
	Dwo	Wa I	) on	Val	Dho	Wal.	Co.	mb ~	บาไ	Nan	Dro	Lou	1	C1	Dwa	Dwa
	PIO	Val	-	Val	File	Val	361		Val	veh	PIO	Deu	-	GIU	PIO	PIO
			355					360					365			
10				_						_						
10	Leu	Val	Thr	Ala	Asn	Thr	Val	Leu	Ser	Ile	Leu	Ser	Val	Asp	Tyr	Pro
		370					375					380				
	Val	Asp	Lys	Val	Ala	Cys	Tyr	Val	Ser	Asp	Asp	Gly	Ser	Ala	Met	Leu
	385					390					395					400
15																
	Thr	Phe	Glu	Ser	Leu	Ser	Glu	Thr	Ala	Glu	Phe	Ala	Lys	Lys	Trp	Val
					405					410					415	
	Pro	Phe	Cvs	Lys	Lvs	Phe	Asn	Ile	Glu	Pro	Arg	Ala	Pro	Glu	Phe	Tvr
20			•	420	•				425					430		
				120					123					-30		
	Dha	21-	C1-	T	710	N am	T	t ou	T	3	*	710	<b>~1</b> -	Desa		Dha
	Pne	ATA		Lys	He	Asp	TYE		rys	Asp	гув	116		PIO	ser	Pne
			435					440					445			
25	Val	ГÀЗ	Glu	Arg	Arg	Ala	Met	Lys	Arg	Glu	Tyr	Glu	Glu	Phe	Lys	Val
		450					455					460				
	Arg	Ile	Asn	Ala	Leu	Val	Ala	Lys	Ala	Gln	Lys	Ile	Pro	Glu	Glu	Gly
	465					470					475					480
30																
	Trp	Thr	Met	Gln	Asp	Gly	Thr	Pro	Trp	Pro	Gly	Asn	Asn	Thr	Arq	qeA
	_				485	_				490	-				495	-
	His	Pro	Glv	Met	Tle	Gln	Val	Phe	Lev	Glv	Hie	Ser	Glv	Glv	T.e.u	Agr
35			01,			02	,,,			o.,		501	Gry	_	neu	vob
,,				500					505					510		
				_			_	_			_		_			
	Tnr	Asp	GIA	Asn	Glu	Leu	Pro	Arg	Leu	He	Tyr	val	Ser	Arg	Glu	Lys
			515					520					525			

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	Arg	Pro	Gly	Phe	Gln	His	His	Lys	Lys	Ala	Gly	Ala	Met	Asn	Ala	Leu
		530					535					540				
	Ile	Arg	Val	Ser	Val	Val	Leu	Thr	Asn	Gly	Ala	Tyr	Leu	Leu	Asn	Val
5	545					550					555					560
	Acn	Cys	Acn	ui e	T) 1.2	Dhe	) cn	Acn	202	Lve	בות	Tla	Tue	C1	71.	Mak
	лор	Cys	veh	nış		FIIC	ASII	non	Ser		nia	116	Lys	GIU		Met
					565					570					575	
10	_					_										
10	Cys	Phe	Met		Asp	Pro	Ala	He		Lys	Lys	СЛВ	Cys	Tyr	Val	Gln
				580					585					590		
	Phe	Pro	Gln	Arg	Phe	Asp	Gly	Ile	Asp	Leu	His	Asp	Arg	Tyr	Ala	Asn
			595					600					605			
15																
	Arg	Asn	Ile	Val	Phe	Phe	Авр	11e	Asn	Met	Lys	Gly	Leu	Asp	Gly	Ile
		610					615					620				
	Gln	Gly	Pro	Val	Tyr	Val	Gly	Thr	Gly	Сув	Cys	Phe	Asn	Arg	Gln	Ala
20	625					630				-	635					640
	Leu	Tyr	Glv	Tvr	Asp	Pro	Val	Leu	Thr	Glu	Glu	Asp	ī,en	Glu	Pro	Asn
		-,-	,	- , -	645					650				<b></b>	655	
					043					050					033	
25	<b>71</b> -	<b>-</b> 1 -		•	• • • •	•	•	-3								
23	iie	Ile	vaı		ser	Сув	Сув	GIY		Arg	Lys	ràs	GIÀ		ser	ser
				660					665					670		
	Lys	Lys	Tyr	Asn	Tyr	Glu	Lys	Arg	Arg	Gly	Ile	Asn	Arg	Ser	Asp	Ser
			675					680					685			
30																
	Asn	Ala	Pro	Leu	Phe	Asn	Met	Glu	Asp	Ile	Asp	Glu	Gly	Phe	Glu	Gly
		690					695					700				
	Tyr	Asp	Asp	Glu	Arg	Ser	Ile	Leu	Met	Ser	Gln	Arg	Ser	Val	Glu	Lys
35	705	_	_			710					715	_				720
	Ara	Phe	glv	G) n	Ser	Pro	V=1	Phe	פוד	Δls	Δls	Thr	Dhe	Met	G1 11	@1 n
	3		1		725					730	-124	• • • •		.100	735	7211
															,,,,	

	Gly	Gly	Ile	Pro	Pro	Thr	Thr	Asn	Pro	Ala	Thr	Leu	Leu	Lys	Glu	Ala
				740					745					750		
	Ile	Hıs	Val	Ile	Ser	Cys	Gly	Tyr	Glu	Asp	Lys	Thr	Glu	Trp	Gly	Lys
5			755					760					765			
	Glu	Ile	Gly	Trp	Ile	Tyr	GJÀ	Ser	Val	Thr	Glu	qaA	Ile	Leu	Thr	Gly
		770					775					780				
10	Phe	Lys	Met	His	Ala		Gly	Trp	Ile	Ser	Ile	Tyr	Сув	Asn	Pro	Pro
	785					790					795					800
	Arg	Pro	Ala	Phe	-	Gly	Ser	Ala	Pro		Asn	Leu	Ser	Asp	_	Leu
15					805					810					815	
13		_,														
	Asn	GIn	Val	Leu	Arg	Trp	Ala	ren	_	Ser	11e	GIU	He		ren	ser
				820					825					830		
	A	ui a	٥.,,	Pro	710	(Tares	Th ess	<b>C1</b>	T	ui o	<i>a</i> 3	N	7 011	7. w.w	Lou	t ou
20	Arg	піз	835	PLO	116	пр	ıyı	840	ıyı	nis	GIY	vra	845	Arg	Dea	Deu
20			633					040					043			
	Glu	Δra	Ile	Ala	Tvr	Tle	Asn	Thr	Tle	Val	Tvr	Pro	alī	Thr	Ser	Tle
	0.0	850		****	-,-		855	••••		•••	-,-	860		****	001	
		030					000									
25	Pro	Leu	Ile	Ala	Tvr	Cvs	Ile	Leu	Pro	Ala	Phe	Cvs	Leu	Ile	Thr	qsA
	865				•	870					875					880
	Arg	Phe	Ile	Ile	Pro	Glu	Ile	Ser	naA	Tyr	Ala	Ser	Ile	Trp	Phe	Ile
					885					890					895	
30																
	Leu	Leu	Phe	Ile	Ser	Ile	Ala	Val	Thr	Gly	Ile	Leu	Glu	Leu	Arg	Trp
				900					905					910		
	Ser	Gly	Val	Ser	Ile	Glu	Asp	Trp	Trp	Arg	Asn	Glu	Gln	Phe	Trp	Val
35			915					920					925			
	Ile	Gly	Gly	Thr	Ser	Ala	His	Leu	Phe	Ala	Val	Phe	Gln	Gly	Leu	Leu
		930					935					940				

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Lys Val Leu Ala Gly Ile Asp Thr Asn Phe Thr Val Thr Ser Lys Ala
945 950 955 960

Thr Asp Glu Asp Gly Asp Phe Ala Glu Leu Tyr Ile Phe Lys Trp Thr 5 965 970 975

Ala Leu Leu Ile Pro Pro Thr Thr Val Leu Leu Val Asn Leu Ile Gly 980 985 990

10 Ile Val Ala Gly Val Ser Tyr Ala Val Asn Ser Gly Tyr Gln Ser Trp
995 1000 1005

Gly Pro Leu Phe Gly Lys Leu Phe Phe Ala Leu Trp Val Ile Ala His 1010 1015 1020

15

Leu Tyr Pro Phe Leu Lys Gly Leu Leu Gly Arg Gln Asn Arg Thr Pro 1025 1030 1035 1040

Thr Ile Val Ile Val Trp Ser Val Leu Leu Ala Ser Ile Phe Ser Leu 20 1045 1050 1055

Leu Trp Val Arg Ile Asn Pro Phe Val Asp Ala Asn Pro Asn Ala Asn 1060 1065 1070

25 Asn Phe Asn Gly Lys Gly Gly Val Phe 1075 1080

- 30 (2) INFORMATION FOR SEQ ID NO:13:
  - (i) SEQUENCE CHARACTERISTICS:

(A) LENGTH: 1741 base pairs

(B) TYPE: nucleic acid

35 (C) STRANDEDNESS: single

(D) TOPOLOGY: linear

- (ii) MOLECULE TYPE: cDNA
- 40 (iii) HYPOTHETICAL: NO

- 163 -

		(iv	AN'	ri-si	ENSE	: NO											
		(vi		IGIN					:								
5			()	A) O	RGAIN.	ISM:	Ory	za s	ativ	<b>a.</b>							
J		י ביע)	MT i	MEDIA	ATE S	SOUR	re.										
				B) CI													
		(ix	) FE	ATURI	Ε:												
10			(2	A) N	AME/I	KEY:	CDS										
			(1	B) L	CAT:	ION:	101	17	41								
		(xi	SE(	QUEN	CE DI	ESCR:	IPTI	ON:	SEQ :	ID N	0:13	:					
15																	
	GTG	CGGC	CGC (	CGCG	CATC	ra G	CTT	CCG	C GC	GCGC	3CGG	ATC	rgcg	AGC	TGCG'	TAGCC	G 60
	TTT	CTCG	CTG !	rgag'	rgga(	GG A	GGAG(	GAGG.	A AG	GGAG	BAGG						115
20												met 1	Ala	Ala	Asn	AIA 5	
20												•				5	
	GGG	ATG	GTG	GCG	GGA	TCC	CGC	AAC	CGG	AAC	GAG	TTC	GTC	ATG	ATC	CGC	163
										Asn							
					10					15					20		
25																	
	ccc	GAC	GGC	GAC	GCG	CCA	CCG	CCG	GCT	AAG	CCA	GGG	AAG	AGT	GTG	AAT	211
	Pro	Asp	Gly	Asp	Ala	Pro	Pro	Pro	Ala	Lys	Pro	Gly	Lys	Ser	Val	Asn	
				25					30					35			
30	GGT																259
	Gly	Gln		Cys	Gln	Ile	Сув	Gly	Asp	Thr	Val	Gly	Val	Ser	Ala	Thr	
			40					45					50				
	000	a.a	ama	mmm	c mm	000	maa		22.0	800							
35	Gly									TGC							307
33	GIY	55	vai	FILE	Val	AId	60 60	ASII	GIU	сув	AIA		PIO	vai	Cys	Arg	
		,,					30					65					
	CCT	TGC	TAC	GAG	TAC	GAA	CGC	AAG	GAA	GGG	AAC	CAG	TGC	TGC	CCC	CAG	359
										Gly							
40	70		•		•	75	,	•		- •	80			4 -		85	

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	TGC	AAG	ACT	AGA	TAC	AAG	AGG	CAC	AAA	GGT	TGC	CCT	AGA	GTT	CAG	GGC	403
	Суѕ	Lys	Thr	Arg	Tyr	Lys	Arg	His	Lys	Gly	Сув	Pro	Arg	Val	Gln	Gly	
					90					95					100		
_																	
5	GAT	GAĢ	GAA	GAA	GAA	GAT	GTT	GAT	GAC	CTG	GAC	TAA	GAA	TTC	CAT	TAT	451
	Asp	Glu	Glu		Glu	Asp	Val	Asp	_	Leu	Asp	Asn	Glu	Phe	His	Tyr	
				105					110					115			
10					GGC												499
IU	Lys	His		Asn	Gly	Lys	Gly		Glu	Trp	Gln	Ile		Arg	Gln	Gly	
			120					125					130				
	CAA	C N TT	CTT	CNC	CTG	THE CHIT	<b>ም</b> ር እ	m~m	m.C∾m	CCC	CAC	CAR	CAA	CNT	ccc	N STATE	547
					Leu												247
15	GIU	135	Val	vob	ucu	361	140	Jer	361	nry	1120	145	<b>J111</b>	nio	nry	116	
••		133					140					117					
	ccc	CGT	CTG	ACA	AGT	GGG	CAA	CAG	ATC	TCA	GGA	GAG	ATC	ССТ	GAT	GCT	595
					Ser												
	150	_				155					160				•	165	
20																	
	TCC	CCC	GAT	CGC	CAT	TCT	ATC	CGC	AGC	GGA	ACA	TCA	AGC	TAT	GTT	GAT	643
	Ser	Pro	Asp	Arg	His	Ser	Ile	Arg	Ser	Gly	Thr	Ser	Ser	Tyr	Val	Asp	
					170					175					180		
25	CCA	AGT	GTT	CCA	GTT	CCT	GTG	AGG	TTA	GTG	GAC	CCC	TCC	AAG	GAC	TTG	691
	Pro	Ser	Val	Pro	Val	Pro	Val	Arg	Ile	Val	Asp	Pro	Ser	Lys	Asp	Leu	
				185					190					195			
• •					ATT									_			739
30	Asn	Ser	Tyr	Gly	Ile	Asn	Ser	Val	Asp	Trp	Gln	Glu		Val	Ala	Ser	
			200					205					210				
									_							TAT	787
25	Trp			Lys	Gln	Asp		Asn	Met	Met	Gln		Ala	Asn	Lys	Tyr	
35		215					220					225					
	CCI	~~~	CC2	NO.	<b>C</b> CC	ccr	<b>a.</b> a	7 m~	an r	ccc	7 Cm		<b>m</b> 0	,,,	-	CDD	025
																GAA	835
		oru	WIG	мg	GIÀ	_	_	Met	GIÜ	GIÀ		GIÀ	ser	ASN	GIĀ	Glu	
40	230					235					240					245	

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	GAT	ATC	CAA	ATG	GTT	GAT	GAT	GCA	CGT	CTA	CCT	CTG	AGC	CGC	ATA	GTG	883
	Asp	Ile	Gln	Met	Val	Asp	Asp	Ala	Arg	Leu	Pro	Leu	Ser	Arg	Ile	Val	
					250					255					260		
_																	
5	CCT	ATC	CCT	TCA	AAC	CAG	CTC	AAC	CTT	TAC	CGG	TTA	GTT	ATC	TTA	CTC	931
	Pro	Ile	Pro	Ser	Asn	Gln	Leu	Asn	Leu	Tyr	Arg	Ile	Val	Ile	Ile	Leu	
				265					270					275			
10				ATC													979
10	Arg	Leu		Ile	Leu	Met	Phe		Phe	Gin	Tyr	Arg		Thr	His	Pro	
			280					285					290				
	CTC	000	CAT	GCT	TAT	CCA	T-TI-C	TCC	CTA	CTA	TICT!	Contra	እ ሞርግ	ጥርጥ	C	N TPTT	1027
				Ala													1027
15	vai	295	Азр	ALA	ıyı	GIY	300	ırp	Leu	vai	361	305	116	Cys	Giu	116	
1.5		2.33					300					303					
	TGG	ттс	CCC	TTA	TCC	TGG	CTC	CTA	GAT	CAA	TTC	CCA	AAG	TGG	TAC	CCG	1075
				Leu													
	310				-	315					320	•	-,-		-,-	325	
20																	
	ATA	AAC	CGT	GAA	ACA	TAC	CTT	GAC	AGG	CTT	GCA	TTG	AGA	TAT	GAT	AGG	1123
	Ile	Asn	Arg	Glu	Thr	Tyr	Leu	Asp	Arg	Leu	Ala	Leu	Arg	Tyr	Asp	Arg	
					330					335					340		
25	GAG	GGA	GAG	CCA	TCA	CAG	CTT	GCT	CCC	ATT	GAT	GTC	TTT	GTC	AGT	ACG	1171
	Glu	Gly	Glu	Pro	Ser	Gln	Leu	Ala	Pro	Ile	Asp	Val	Phe	Val	Ser	Thr	
				345					350					355			
	GTG	GAT	CCA	CTA	AAG	GAA	CCT	CCT	CTG	ATC	ACA	GCA	AAC	ACT	GTT	TTG	1219
30	Val	Asp	Pro	Leu	Lys	Glu	Pro	Pro	Leu	Ile	Thr	Ala	Asn	Thr	Val	Leu	
			360					365					370				
	TCC	ATT	CTG	GCT	GTG	GAT	TAC	CCT	GTT	GAC	AAA	GTG	TCA	TGC	TAT	GTT	1267
	Ser	Ile	Leu	Ala	Val	Asp	Tyr	Pro	Val	Asp	ГÀв	Val	Ser	Сув	Tyr	Val	
35		375					380					385					
				GGT	-												1315
		Asp	Asp	Gly	Ser		Met	Leu	Thr	Phe		Ala	Leu	Ser	Glu		
40	390					395					400					405	
70																	

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	GCA	GAA	TTT	GCT	AGG	AAG	TGG	GTT	CCG	TTT	TGC	AAG	AAG	CAC	AAT	ATT	1363
	Ala	Glu	Phe	Ala	Arg	Lys	Trp	Val	Pro	Phe	Cys	Lys	Lys	His	Asn	Ile	
					410					415					420		
5	gaa	CCA	CGA	GCT	CCA	GAG	TTT	TAC	TTT	GCT	CAA	AAA	ATA	GAT	TAC	CTG	1411
	Glu	Pro	Arg	Ala	Pro	Glu	Phe	Tyr	Phe	Ala	Gln	Lys	Ile	Asp	Tyr	Leu	
				425					430					435			
					CAA												1459
10	Lys	Aap	Lys	Ile	Gln	Pro	Ser	Phe	Val	Lys	Glu	Arg	Arg	Ala	Met	Lys	
			440					445					450				
	AGA	GAG	TAT	gaa	GAA	TTC	AAG	GTA	CGG	ATC	AAT	GCT	CTT	GTT	GCG	AAG	1507
	Arg	Glu	Tyr	Glu	Glu	Phe	Lys	Val	Arg	Ile	Asn	Ala	Leu	Val	Ala	Lys	
15		455					460					465					
	GCA	CAA	AAA	GTA	CCT	GAA	GAG	GGG	TGG	ACC	ATG	GCT	GAT	GGC	ACT	GCT	1555
	Ala	Gln	ràa	Val	Pro	Glu	Glu	Gly	Trp	Thr	Met	Ala	Aap	Gly	Thr	Ala	
••	470					475					480					485	
20																	
	TGG	CCT	GGG	AAT	AAC	CCA	AGG	GAT	CAC	CCT	GGC	ATG	ATT	CAG	GTG	TTC	1603
	Trp	Pro	Gly	Asn	Asn	Pro	Arg	Asp	His	Pro	Gly	Met	Ile	Gln	Val	Phe	
					490					495					500		
25																	
23					GGT												1651
	Leu	Gly	His		Gly	Gly	Leu	qaA		Asp	Gly	Asn	Glu		Pro	Arg	
				505					510					515			
20					TCT												1699
30	Leu	Val		Val	Ser	Arg	Glu		Arg	Pro	Gly	Phe		His	His	Lys	
			520					525					530				
						• • •											
					ATG												1741
25	rys		Gly	Ala	Met	Asn		Leu	He	Arg	Val		Ala	Val			
35		535					540					545					

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	(2)	INF	ORMAT	поі	FOR	SEQ	ID I	NO:14	<b>1</b> :							
			(i) S	SEQUI	ENCE	CHAI	RACTI	ERIST	rics	:						
				(A)	LE	NGTH:	54	7 am:	ino a	acids	S					
5				(B)	TY	PE: a	mino	ac	id							
				(D)	TO	POLO	3Y: ]	linea	ar							
		(:	11) N	OLE	CULE	TYPI	E: p	rote:	in							
10		(;	ki) S	SEQUI	ENCE	DES	CRIP:	rion	: SE(	) ID	NO:	14:				
	Met	Ala	Ala	Asn	Ala	Gly	Met	Val	Ala	Gly	Ser	Arg	Asn	Arg	Asn	Glu
	1				5					10					15	
15	Phe	Val	Met	Ile	Arg	Pro	Asp	Gly	Asp	Ala	Pro	Pro	Pro	Ala	Lys	Pro
				20					25					30		
	Gly	Lys	Ser	Val	Asn	Gly	Gln	Val	Сув	Gln	Ile	Сув	Gly	Asp	Thr	Val
			35					40					45			
20																
	Gly	Val	Ser	Ala	Thr	Gly	Asp	Val	Phe	Val	Ala	Сув	Asn	Glu	Сув	Ala
		50					55					60				
•	Phe	Pro	Val	Cys	Arg	Pro	Сув	Tyr	Glu	Tyr	Glu	Arg	Lys	Glu	Gly	Asn
25	65					70					75					80
	Gln	Cys	Cys	Pro		Cys	Lys	Thr	Arg	Tyr	Lys	Arg	His	Lys	_	Cys
					85					90					95	
20	_					_							_			
30	Pro	Arg	Val		Gly	Asp	Glu			Glu	Asp	Val	Asp	qaA	Leu	Asp
				100					105					110		
	N	G)	nh -	***	<b></b>	•	,,,, _	<b>a</b> 3		<b>03</b>	*	<b>03</b>	<b>5</b>	<b>~1</b>	<b></b>	<b>01</b>
	ASN	GIU	Phe	HIS	ıyr	rys	HIS		Asn	GIA	гÀв	GIÀ		Glu	Trp	GIn
35			115					120					125			
33			_							_	_	_		_		
	He		Arg	GIn	Gly	Glu	-	Val	Asp	Leu	Ser		Ser	Ser	Arg	His
		130					135					140				
	۵,	٠,				_		_		_					_	
<b>4</b> 0	Glu	Gln	His	Arg			Arg	Leu	Thr	Ser		Gln	Gln	Ile	Ser	
	1/15					150					1 5 5					160

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	Glu	Ile	Pro	Asp		Ser	Pro	Asp	Arg		Ser	Ile	Arg	Ser	_	Thr
					165					170					175	
	Ser	Ser	туг	Val	Asp	Pro	Ser	Val	Pro	Val	Pro	Val	Arg	Ile	Val	Asp
5				180					185					190		
	Pro	Ser	Lys	Asp	Leu	Asn	Ser	Tyr	Gly	Ile	Asn	Ser	Val	Asp	Trp	Gln
			195					200					205			
10	<b>61</b>	2	1/01	Ala	C - =	<b></b>	D	۸	T = ==	C) =	N ===		Non.	Man	W-+	<b>61</b>
10	GIU	210	VAI	AIS	ser	IIP	215	ASII	гув	GIII	Aab	220 220	Asn	Mec	Met	GIN
		Ala	Asn	Lys	Tyr		Glu	Ala	Arg	Gly		Asp	Met	Glu	Gly	
15	225					230					235					240
	Gly	Ser	Asn	Gly	Glu	Asp	Ile	Gln	Met	Val	Asp	Asp	Ala	Arg	Leu	Pro
					245					250					255	
	Leu	Ser	Arg	Ile	Val	Pro	Ile	Pro	Ser	Asn	Gln	Leu	Asn	Leu	Tyr	Arg
20				260					265					270		
	Tle	Val	Tle	Ile	Leu	Ara	Len	Tle	Tle	Len	Met	Phe	Dhe	Phe	Gln	Tur
	110	vaz	275	110	Deu	Arg	#ea	280	116	пси	Mec	File	285	FIIC	JI.	
25	Arg	Val 290	Thr	His	Pro	Val	Arg 295	Asp	Ala	Tyr	Gly	Leu 300	Trp	Leu	Val	Ser
		230					233					300				
	Val	Ile	Сув	Glu	Ile	Trp	Leu	Pro	Leu	Ser	Trp	Leu	Leu	Asp	Gln	Phe
30	305					310					315					320
50	Pro	Lys	Trp	Tyr	Pro	Ile	Asn	Arg	Glu	Thr	Tyr	Leu	Asp	Arg	Leu	Ala
					325					330					335	
	7.00	<b>&gt;</b>	<b></b>	<b>&gt;</b>	2	<b>23</b>	<b>61</b>	<b>63.</b>	Duna	C	<b>61</b> -	• • • •	21-	D	77	
35	reu	Arg	ıyı	Asp 340	Arg	GIU	GIY	GIU	345	ser	GIII	Leu	Ald	350	ire	Авр
	Val	Phe		Ser	Thr	Val	Asp		Leu	Lys	Glu	Pro		Leu	Ile	Thr
			355					360					365			

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	Ala	Asn	Thr	Val	Leu	Ser	Ile	Leu	Ala	Val	Asp	Tyr	Pro	Val	Asp	Lys
		370					375					380				
5	Val	Ser	Cvs	Tyr	Val	Ser	Asp	Asp	Glv	Ser	Ala	Met	Leu	Thr	Phe	Glu
•	385		-7-	-,-		390			•		395					400
	303					330					,,,					400
					0					_	_	_		_		_
	Ala	Leu	Ser	Glu	Thr	Ala	Glu	Phe	Ala	-	Lys	Trp	Val	Pro		Cys
					405					410					415	
10																
	Lys	Lys	His	Asn	Ile	Glu	Pro	Arg	Ala	Pro	Glu	Phe	Tyr	Phe	Ala	Gln
				420					425					430		
	Lvs	tle	Asp	Tyr	Leu	Lvs	Asp	Lvs	Ile	Gln	Pro	Ser	Phe	Val	Lvs	Glu
15	-,-		435	-7-		-,-		440					445		•	
13			433					440					***			
								_				_		_		
	Arg	Arg	Ala	Met	Lys	Arg	Glu	Tyr	Glu	Glu	Phe	Lys	Val	Arg	Ile	Asn
		450					455					460				
20	Ala	Leu	Val	Ala	Lys	Ala	Gln	Lys	Val	Pro	Glu	Glu	Gly	Trp	Thr	Met
	465					470					475					480
	a [ 4	Aen	Gly	Thr	Δla	Trn	Pro	Glv	Agn	Agn	Pro	Ara	Asp	His	Pro	Glv
	n.u	rap	O.,	****	485			<b>0.</b> ,		490		••••	···		495	,
25					403					430					422	
23																
	Met	Ile	Gln	Val	Phe	Leu	Gly	His	Ser	Gly	Gly	Leu	Asp	Thr	qaA	Gly
				500					505					510		
	Asn	Glu	Leu	Pro	Arg	Leu	Val	Tyr	Val	Ser	Arg	Glu	Lys	Arg	Pro	Gly
30			515					520					525			
	Dhe	Gln	uie	His	Lva	Lara	Ala	Gly	Δla	Met	Aan	Δla	T.em	Tle	Ara	Val
	FIIE			นาย	ոչ	ъyв		GTA	viq	rie C	VOII		Ter	*16	A. y	VQI
		530					535					540				
35	Ser	Ala	Val													
	545															

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## **CLAIMS:**

- An isolated nucleic acid molecule which encodes a polypeptide of the cellulose biosynthetic pathway or a homologue, analogue or derivative thereof or a complementary sequence thereto, wherein said polypeptide is capable of producing cellulose and/or β-1,4-5 glucan and/or an intermediate between cellulose and a β-1,4-glucan polymer.
  - 2. The isolated nucleic acid molecule according to claim 1 wherein the polypeptide is cellulose synthase or a catalytic subunit thereof.
- 10 3. The isolated nucleic acid molecule according to claim 1 or 2, derived from a prokaryote.
- 4. The isolated nucleic acid molecule according to claim 3, wherein the prokaryote is a bacterium other than Agrobacterium tumefaciens. Acetobacter pasteurianus or Acetobacter 15 xylinum.
  - 5. The isolated nucleic acid molecule according to claim 1 or 2, derived from a eukaryote.
- 20 6. The isolated nucleic acid molecule according to claim 5, wherein the eukaryote is a plant or fungus.
  - 7. The isolated nucleic acid molecule according to claim 6, wherein the plant is selected from the list comprising *Arabidopsis thaliana*, *Gossypium hirsutum* (cotton), *Oryza sativa*
- 25 (rice), wheat, barley, maize, Brassica ssp., Eucalyptus ssp., hemp, jute, flax, Pinus ssp., Populus ssp., and Picea spp., amongst others.
  - 8. The isolated nucleic acid molecule according to claim 2 wherein the cellulose synthase or catalytic subunit thereof is the *Arabidopsis thaliana* RSW1 polypeptide.

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9. The isolated nucleic acid molecule according to any one of claims 1 to 8, comprising a sequence of nucleotides which is at least 40% identical to any one of SEQ ID NOs:1, 3, 4, 5, 7, 9, 11 or 13 or a complementary sequence thereof.

- 10. The isolated nucleic acid molecule according to claim 9, wherein the percentage identity to any one of SEQ ID NOs:1, 3, 4, 5, 7, 9, 11 or 13 or a complementary sequence thereof is at least 60%.
- 10 11. The isolated nucleic acid molecule according to claim 9, wherein the percentage identity to any one of SEQ ID NOs:1, 3, 4, 5, 7, 9, 11 or 13 or a complementary sequence thereof is at least 80%.
- 12. An isolated nucleic acid molecule which comprises a sequence of nucleotides substantially as set forth in any one of SEQ ID NOs:3, 4, 5, 7, 9 or 11 or a homologue, analogue or derivative thereof or a complementary sequence thereto.
- 13. The isolated nucleic acid molecule according to any one of claims 1 to 12, wherein said nucleic acid molecule hybridizes under at least low stringency conditions to at least 20 contiguous nucleotides of any one of SEQ ID NOs:1, 3, 4, 5, 7, 9, 11 or 13 or a complementary sequence thereto.
- 14. An isolated nucleic acid molecule which encodes a polypeptide which is capable of cellulose and/or β-1,4- glucan biosynthesis in a plant cell, fungal cell, insect cell. animal cell,
  25 yeast cell or bacterial cell when expressed therein.
  - 15. The isolated nucleic acid molecule according to claim 14, wherein the polypeptide is cellulose synthase or a catalytic subunit thereof.
- 30 16. The isolated nucleic acid molecule according to claim 14 or 15, derived from a

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prokaryote.

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- 17. The isolated nucleic acid molecule according to claim 16, wherein the prokaryote is a bacterium other than Agrobacterium tumefaciens, Acetobacter pasteurianus or Acetobacter 5 xylinum.
  - 18. The isolated nucleic acid molecule according to claim 14 or 15, derived from a eukaryote.
- 10 19. The isolated nucleic acid molecule according to claim 18, wherein the eukaryote is a plant or fungus.
- The isolated nucleic acid molecule according to claim 19, wherein the plant is selected from the list comprising Arabidopsis thaliana, Gossypium hirsutum (cotton), Oryza sativa
  (rice), wheat, barley, maize, Brassica ssp., Eucalyptus ssp., hemp, jute, flax, Pinus ssp., Populus ssp., and Picea spp., amongst others.
  - 21. The isolated nucleic acid molecule according to claim 20, wherein the cellulose synthase or catalytic subunit thereof is the *Arabidopsis thaliana* RSW1 polypeptide.

22. The isolated nucleic acid molecule according to any one of claims 14 to 21, comprising a sequence of nucleotides which is at least 40% identical to any one of SEQ ID NOs:1, 3, 4, 5, 7, 9, 11 or 13 or a complementary sequence thereto.

- 25 23. The isolated nucleic acid molecule according to claim 22, wherein the percentage identity to any one of SEQ ID NOs:1, 3, 4, 5, 7, 9, 11 or 13 or a complementary sequence thereof is at least 60%.
- 24. The isolated nucleic acid molecule according to claim 22, wherein the percentage 30 identity to any one of SEQ ID NOs:1, 3, 4, 5, 7, 9, 11 or 13 or a complementary sequence

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thereof is at least 80%.

- 25. The isolated nucleic acid molecule according to claim 22, comprising the sequence of nucleotides substantially as set forth in any one of SEQ ID NOs:3, 4, 5, 7, 9 or 11 or a 5 homologue, analogue or derivative thereof or a complementary sequence thereto.
- 26. An isolated nucleic acid molecule which encodes or is complementary to a nucleic acid molecule which encodes a polypeptide capable of cellulose and/or β-1.4-glucan biosynthesis wherein said polypeptide comprises a sequence of amino acids which is at least
  10 40% identical to any one of SEQ ID Nos:2, 6, 8, 10, 12 or 14.
  - 27. The isolated nucleic acid molecule according to claim 26, wherein the percentage identity to any one of SEQ ID Nos:2, 6, 8, 10, 12 or 14 is at least 60%.
- 15 28. The isolated nucleic acid molecule according to claim 27, wherein the percentage identity to any one of SEQ ID Nos:2, 6, 8, 10, 12 or 14 is at least 80%.
- 29. The isolated nucleic acid molecule according to claim 26, wherein the polypeptide comprises a sequence of amino acids substantially as set forth in any one of SEQ ID Nos:2, 20 6, 8, 10, 12 or 14.
  - 30. A genetic construct which comprises the isolated nucleic acid molecule according to any one of claims 1 to 29.
- 25 31. A genetic construct which comprises the isolated nucleic acid molecule according to any one of claims 1 to 29 operably connected to a promoter sequence.
- 32. The genetic construct according to claim 31, wherein the nucleic acid molecule is operably connected to the promoter sequence in the sense orientation such that RNA which 30 encodes a polypeptide capable of cellulose and/or β-1,4-glucan biosynthesis or a homologue,

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analogue or derivative thereof is produced when said nucleic acid molecule is expressed.

- 33. The genetic construct according to claim 31, wherein the nucleic acid molecule is operably connected to the promoter sequence in the antisense orientation such that RNA which is complementary to RNA which encodes a polypeptide capable of cellulose and/or β-1,4-glucan biosynthesis or a homologue, analogue or derivative thereof, is produced when said nucleic acid molecule is expressed.
- 34. The genetic construct according to claim 33, wherein the nucleic acid molecule 10 encodes an antisense or ribozyme molecule.
  - 35. The genetic construct according to any one of claims 31 to 34, wherein the promoter is the CaMV 35S promoter.
- 15 36. The genetic construct according to any one of claims 31 to 34, wherein the promoter is the *Arabidopsis thaliana RSW*1 gene promoter.
- 37. A method of increasing the level of cellulose in a cell, tissue, organ or organism, said method comprising expressing the isolated nucleic acid molecule according to any one of 20 claims 1 to 29 therein, in the sense orientation, for a time and under conditions at least sufficient to produce or increase expression of the polypeptide encoded therefor.
  - 38. The method according to claim 37, comprising the additional first step of transforming the cell, tissue, organ or organism with the isolated nucleic acid molecule.
  - 39. The method according to claim 38, wherein the cell is a prokaryotic cell.
  - 40. The method according to claim 38, wherein the cell, tissue, organ or organism is a eukaryotic cell, tissue, organ or organism.

- 41. The method according to claim 40, wherein the cell, tissue, organ or organism is a plant, fungal, insect, animal or yeast cell, tissue, organ or organism.
- 42. The method according to claim 41, wherein the cell, tissue, organ or organism is a 5 plant cell, tissue, organ or organism.
- 43. The method according to claim 42 wherein the plant is selected from the list comprising Arabidopsis thaliana, Gossypium hirsutum (cotton), Oryza sativa (rice), Eucalyptus ssp., Brassica ssp., wheat, barley, maize, hemp, jute, flax, and woody plants 10 such as Pinus ssp., Populus ssp., Picea spp., amongst others.
- 44. A method of reducing the level of non-crystalline β-1,4-glucan in a cell, tissue, organ or organism, said method comprising expressing the isolated nucleic acid molecule according to any one of claims 1 to 29 therein, in the sense orientation, for a time and under conditions
  15 at least sufficient to produce or increase expression of the polypeptide encoded therefor.
  - 45. The method according to claim 44, comprising the additional first step of transforming the cell, tissue, organ or organism with the isolated nucleic acid molecule.
  - 46. The method according to claim 44, wherein the cell is a prokaryotic cell.

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- 47. The method according to claim 44, wherein the cell, tissue, organ or organism is a eukaryotic cell, tissue, organ or organism.
- 48. The method according to claim 47, wherein the cell, tissue, organ or organism is a plant, fungal, insect, animal or yeast cell, tissue, organ or organism.
- 49. The method according to claim 48, wherein the cell, tissue, organ or organism is a 30 plant cell, tissue, organ or organism.

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50. The method according to claim 50 wherein the plant is selected from the list comprising Arabidopsis thaliana, Gossypium hirsutum (cotton), Oryza sativa (rice), Eucalyptus ssp., Brassica ssp., wheat, barley, maize, hemp, jute, flax, and woody plants such as Pinus ssp., Populus ssp., Picea spp., amongst others.

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51. A method of reducing the level of starch in a cell, tissue, organ or organism, said method comprising expressing the isolated nucleic acid molecule according to any one of claims 1 to 29 therein, in the sense orientation, for a time and under conditions at least sufficient to produce or increase expression of the polypeptide encoded therefor.

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- 52. The method according to claim 50, comprising the additional first step of transforming the cell, tissue, organ or organism with the isolated nucleic acid molecule.
- 53. The method according to claim 51, wherein the cell is a prokaryotic cell.

- 54. The method according to claim 53, wherein the cell, tissue, organ or organism is a eukaryotic cell, tissue, organ or organism.
- 55. The method according to claim 54, wherein the eukaryote is a plant. fungus, insect, 20 animal or yeast.
  - 56. The method according to claim 55, wherein the eukaryote is a plant.
- 57. The method according to claim 56 wherein the plant is selected from the list comprising Arabidopsis thaliana, Gossypium hirsutum (cotton), Oryza sativa (rice), Eucalyptus ssp., Brassica ssp., wheat, barley, maize, hemp, jute, flax, and woody plants such as Pinus ssp., Populus ssp., Picea spp., amongst others.
- 58. A method of reducing the level of cellulose in a cell, tissue, organ or organism, said 30 method comprising expressing the isolated nucleic acid molecule according to any one of

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claims 1 to 29 therein, in the antisense orientation, for a time and under conditions at least sufficient to prevent or reduce the expression of the polypeptide encoded therefor.

- 59. The method according to claim 58, comprising the additional first step of transforming the cell, tissue, organ or organism with the isolated nucleic acid molecule.
  - 60. The method according to claims 58 or 59, wherein the cell, tissue, organ or organism is a eukaryotic cell, tissue, organ or organism.
- 10 61. The method according to claim 60, wherein the eukaryote is a plant, fungus, insect, animal or yeast.
  - 62. The method according to claim 61, wherein the eukaryote is a plant.
- 15 63. The method according to claim 62 wherein the plant is selected from the list comprising Arabidopsis thaliana, Gossypium hirsutum (cotton), Oryza sativa (rice), Eucalyptus ssp., Brassica ssp., wheat, barley, maize, hemp, jute, flax, and woody plants such as Pinus ssp., Populus ssp., Picea spp., amongst others.
- 20 64. A method of increasing the level of non-crystalline β-1,4-glucan in a cell, tissue, organ or organism, said method comprising expressing the isolated nucleic acid molecule according to any one of claims 1 to 29 therein, in the antisense orientation, for a time and under conditions at least sufficient to prevent or reduce the expression of the polypeptide encoded therefor.

- 65. The method according to claim 64, comprising the additional first step of transforming the cell, tissue, organ or organism with the isolated nucleic acid molecule.
- 66. The method according to claims 64 or 65, wherein the cell, tissue, organ or organism 30 is a eukaryotic cell, tissue, organ or organism.

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- 67. The method according to claim 66, wherein the eukaryote is a plant, fungus, insect, animal or yeast.
- 68. The method according to claim 67, wherein the eukaryote is a plant.

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69. The method according to claim 68 wherein the plant is selected from the list comprising Arabidopsis thaliana, Gossypium hirsutum (cotton), Oryza sativa (rice), Eucalyptus ssp., Brassica ssp., wheat, barley, maize, hemp, jute, flax, and woody plants such as Pinus ssp., Populus ssp., Picea spp., amongst others.

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70. A method of increasing the level of starch in a cell, tissue, organ or organism, said method comprising expressing the isolated nucleic acid molecule according to any one of claims 1 to 29 therein, in the antisense orientation, for a time and under conditions at least sufficient to prevent or reduce the expression of the polypeptide encoded therefor.

- 71. The method according to claim 70, comprising the additional first step of transforming the cell, tissue, organ or organism with the isolated nucleic acid molecule.
- 72. The method according to claims 70 or 71, wherein the cell, tissue, organ or organism 20 is a eukaryotic cell, tissue, organ or organism.
  - 73. The method according to claim 72, wherein the eukaryote is a plant, fungus, insect, animal or yeast.
- 25 74. The method according to claim 73, wherein the eukaryote is a plant.
- 75. The method according to claim 74 wherein the plant is selected from the list comprising Arabidopsis thaliana, Gossypium hirsutum (cotton), Oryza sativa (rice), Eucalyptus ssp., Brassica ssp., wheat, barley, maize, hemp, jute, flax, and woody plants 30 such as Pinus ssp., Populus ssp., Picea spp., amongst others.

- 76. A method of producing a recombinant enzymatically active polypeptide which is capable of synthesizing cellulose and/or β-1,4-glucan and/or an intermediate between cellulose and β-1,4-glucan in a cell, said method comprising expressing the isolated nucleic acid molecule according to any one of claims 1 to 29 or a homologue, analogue or derivative thereof in said cell for a time and under conditions sufficient for the polypeptide encoded therefor to be produced.
- 77. The method according to claim 76, comprising the additional first step of transforming the cell with the isolated nucleic acid molecule according to any one of claims 10 1 to 29 or the genetic construct according to any one of claims 11 to 15.
  - 78. A recombinant polypeptide produced according to the method defined by claim 76 or 77.
- 15 79. The recombinant cellulose biosynthetic polypeptide according to claim 78, further defined as a recombinant cellulose synthase or catalytically active subunit thereof.
- 80. A recombinant cellulose biosynthetic polypeptide capable of cellulose and/or β-1,4-glucan production and comprising a sequence of amino acids set forth in any one of SEQ ID
  20 Nos: 2, 6, 8, 10, 12 or 14 or a homologue, analogue or derivative thereof which is at least 40% identical thereto.
  - 81. The recombinant cellulose biosynthetic polypeptide according to claim 80, wherein the percentage identity to any one of SEQ ID Nos: 2, 6, 8, 10, 12 or 14 is at least 60%.

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- 82. The recombinant cellulose biosynthetic polypeptide according to claim 81, wherein the percentage identity to any one of SEQ ID Nos: 2, 6, 8, 10, 12 or 14 is at least 80%.
- 83. The recombinant cellulose biosynthetic polypeptide according to claim 82, comprising a sequence of amino acids substantially as set forth in any one of SEQ ID Nos: 2, 6, 8, 10,

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12 or 14.

- 84. A method of altering the mechanical properties of a cell wall, said method comprising expressing the isolated nucleic acid molecule according to any one of claims 1 to 29 in the
  5 antisense orientation in said cell for a time and under conditions sufficient for the level of non-crystalline β-1,4-glucan to increase in said cell.
  - 85. The method according to claim 84 wherein the non-crystalline  $\beta$ -1,4-glucan is cross-linked to cellulose microfibrils.

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- 86. The method according to claim 84 or 85 wherein the cell wall normally has a high ratio of cellulose to hemicelluloses.
- 87. The method according to any one of claims 84 to 86, wherein the nucleic acid15 molecule expressed in the antisense orientation is contained within an antisense molecule or ribozyme molecule.
  - 88. The method according to any one of claims 84 to 87, wherein the cell wall is a plant cell wall.

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89. The method according to claim 88, wherein the plant is selected from the list comprising Arabidopsis thaliana, Gossypium hirsutum (cotton), Oryza sativa (rice), Eucalyptus ssp., Brassica ssp., wheat, barley, maize, hemp, jute, flax, and woody plants such as Pinus ssp., Populus ssp., Picea spp., amongst others.

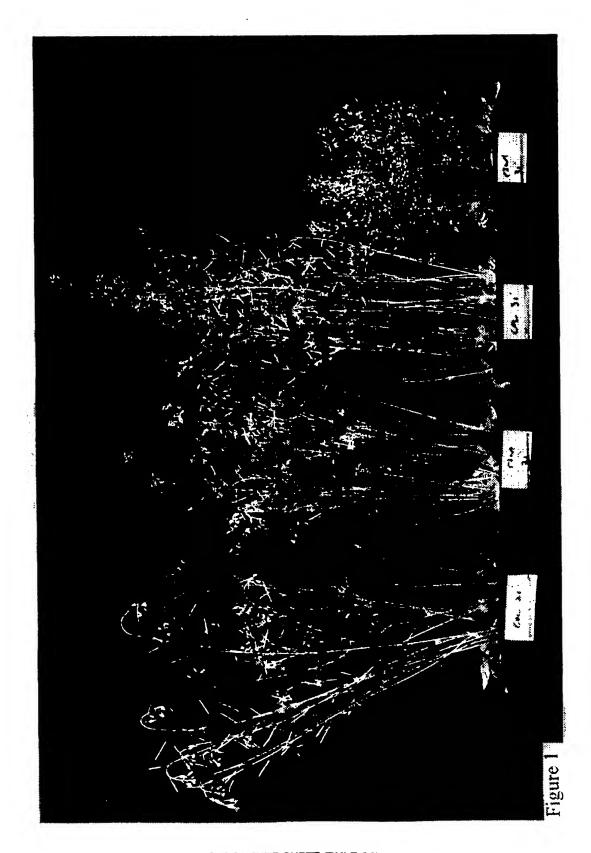
25

- 90. An antibody molecule which binds to the recombinant polypeptide according to any one of claims 78 to 83 or a homologue, analogue or derivative thereof.
- 91. A transgenic plant transformed with the isolated nucleic acid molecule according to 30 any one of claims 1 to 29 or a genetic construct according to any one of claims 30 to 36.

92. The transgenic plant according to claim 91, wherein said plant is selected from the list comprising Arabidopsis thaliana. Gossypium hirsutum (cotton), Oryza sativa (rice), Eucalyptus ssp., Brassica ssp., wheat, barley, maize, hemp, jute, flax, and woody plants such as Pinus ssp., Populus ssp., Picea spp., amongst others.

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- 93. Use of an isolated nucleic acid molecule according to any one of claims 1 to 29 to modify the cellulose content of a cell.
- 94. Use according to claim 93, wherein if the nucleic acid molecule according to any one 10 of claims 1 to 29 is expressed in the sense orientation in said cell, the level of cellulose therein is increased.
- 95. Use according to claim 93, wherein if the nucleic acid molecule according to any one of claims 1 to 29 is expressed in the antisense orientation in said cell, the level of cellulose 15 therein is decreased.
  - 96. Use according to claim 95, wherein said cell is further characterised by increased non-crystalline  $\beta$ -1,4-glucan content and/or starch content.
- 20 97. Use according to claim 95 or 96, wherein said cell is further characterised by increased cross-linking of non-crystalline β-1,4-glucan to cellulose.
  - 98. Use according to any one of claims 93 to 97, wherein the cell is a plant cell.
- 25 99. Use according to claim 98 wherein the plant is selected from the list comprising Arabidopsis thaliana, Gossypium hirsutum (cotton), Oryza sativa (rice), Eucalyptus ssp., Brassica ssp., wheat, barley, maize, hemp, jute, flax, and woody plants such as Pinus ssp., Populus ssp., Picea spp., amongst others.



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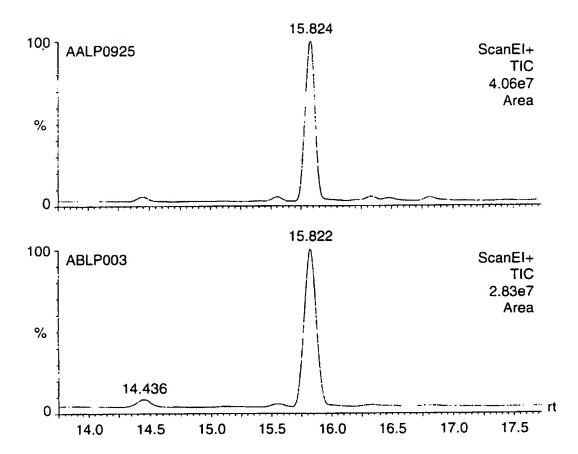


Figure 3

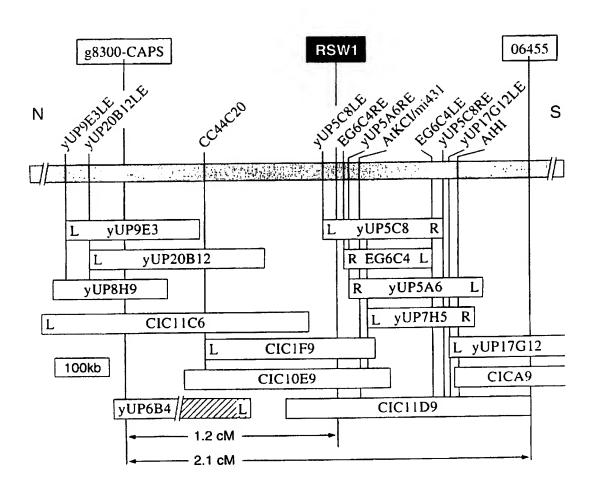


Figure 4

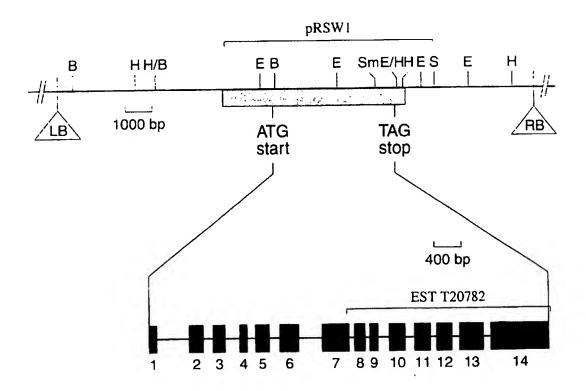
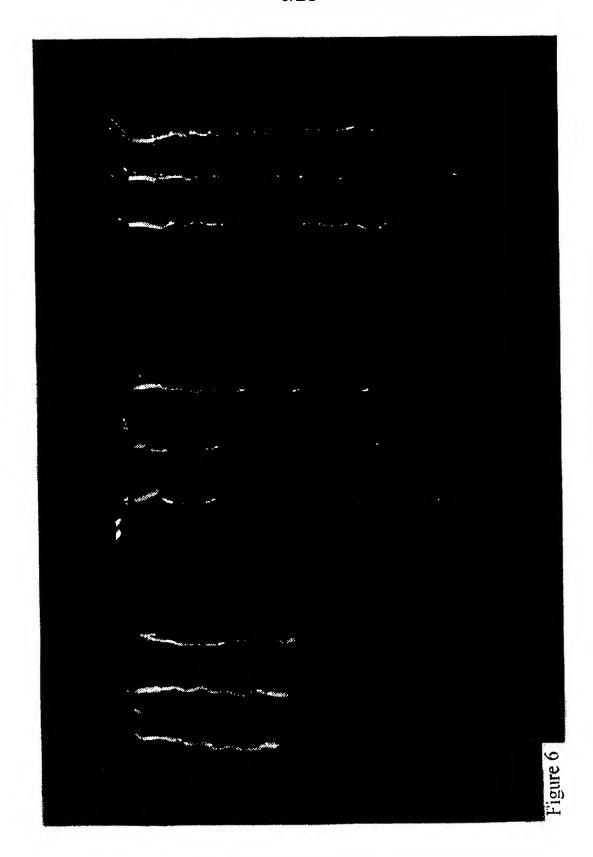
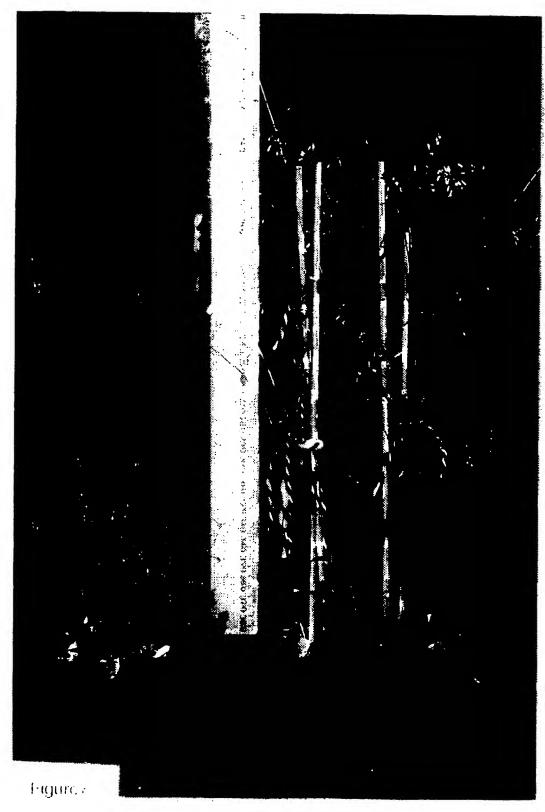


Figure 5



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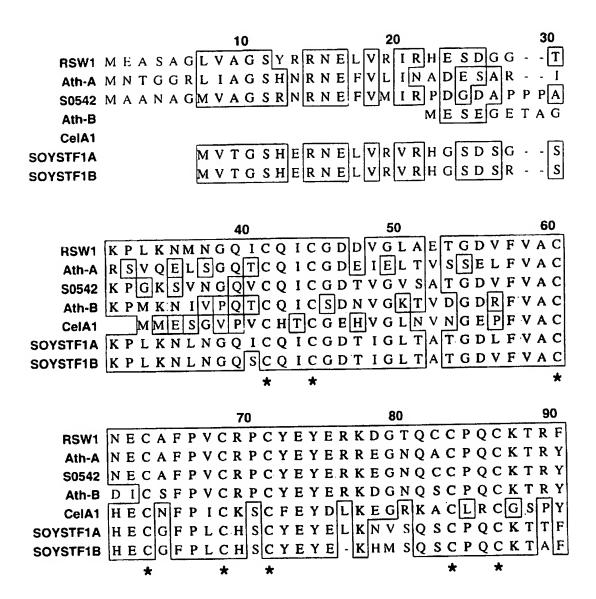


Figure 8

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Cont I Cont II Cont II Cont IV Cont V Cont VI Cont VII Cont VIII Cont IX Cont X

09 120 **NECAFPVCRPCYEYERKEGNQCCPQCKTRYKRHKGCPRVQGDEEEEDVDDLDNEFHYKHG** DICSFPVCRPCYEYERKDGNQSCPQCKTRYKRLKGSPAIPGDKDEDGLADEGTVEFNYPQ MEASAGLVAGSYRRNELVRIRHESDGG..TKPLKNMNGQICQICGDDVGLAETGDVFVAC MNTGGRLIAGSHNRNEFVLINADESAR..IRSVQELSGQTCQICGDEIELTVSSELFVAC **MAANAGMVAGSRNRNEFVMIRPDGDAPPPAKPGKSVNGQVCQICGDTVGVSATGDVFVAC** MESEGETAGKPMKNIVPQTCQICSDNVGKTVDGDRFVAC MMESGVPVCHTCGEHVGLNVNGEPFVAC **NECAFPVCRPCYEYERKDGTQCCPQCKTRFRRHRGSPRVEGDEDDDVDD1ENEFNYAQG NECAFPVCRPCYEYERREGNQACPQCKTRYKRIKGSPRVDGDDEEEEDIDDLEYEFDHGM** HECNFPICKSCFEYDLKEGRKACLRCGSPYDENLLDDVEKATGDQSTMAAHLNKSQDVGI 50 100 90 80 **D48636** 048636 Ath-A Ath-B Ath-A 50542 Ath-B CelA2 S0542 celA1 CelA2 CelA1 RSW1 RSW1

FIGURE 9 (CONT I

	130	140	150	160	170	180
RSW1	ANKA	.RHQRHGEEFS	SSSRHESQP	PLLTHGHTV	RHQRHGEEFSSSRHESQPIPLLTHGHTVSGEIRTPDTQSVRTT	RTT
Ath-A	DPEHAAEAALSSRLNTGRGGLDSAPPGSQIPLLTYCDEDADMYSDRHALIVPPSTGYGNR	<b>TGRGGLDSAPF</b>	GSOIPLLTY	CDEDADMYSDI	RHALIVPPSTG	GNR
S0542	NGKGPEWQI	.QRQGEDVDLS	SSSRHEQHR	PRLTSGQQI	QRQGEDVDLSSSSRHEQHRIPRLTSGQQISGEIPDASPDRHSIR	SIR
Ath-B	K. EKISERMLGWHLTRGKGEEMGEPQYDKEVSHNHLPRLTSRQDTSGEFSAASPERLSVS	<b>TRGKGEEMGEPQ</b>	YDKEVSHNHI	PRLTSRODT	SGEFSAASPERI	SVS
Cel-A1	•			•	•	
Cel-A2						
D48636						
	190	200	210	220	230	240
RSW1	SGPLGPSDRNAISSI	YIDPRQPVPVF	LIVDPSKDLN	SYGLGNVDWK	ISSPYIDPRQPVPVRIVDPSKDLNSYGLGNVDWKERVEGWKLKQEKNML	NML
Ath-A	VYPAI	PFTDSSAPPQAF	RSMVPQKDIA	<b>EYGYGSVAWK</b>	. PAPFTDSSAPPQARSMVPQKDIAEYGYGSVAWKDRMEVWKRRQGEKLQ	KLQ
S0542		SYVDPSVPVPVF	LIVDPSKDLN	SYGINSVDWQ	. TSSYVDPSVPVRIVDPSKDLNSYGINSVDWQERVASWRNKQDKNMM	NMM
Ath-B		GKKLPYSSDVN	<b>IQSPNRRIVD</b>	PVGLGNVAWK	. IAGGKRLPYSSDVNQSPNRRIVDPVGLGNVAWKERVDGWKMKQEKNTG	NTG
Cel-A1		HARHIS	SSVSTLDSEM	AEDNGNS I WKI	HARHISSVSTLDSEMAEDNGNSIWKNRVESWKEKKNKKKK	KKK
Cel-A2						
D48636			S	<b>L'TRPGNVAWK</b>	STTRPGNVAWKERVDGWKLKQDKGAI	GAI
FIGURE	9 (CONT II)					

		250	260	270	280	290	300
RSW1	OMT	GKYHEGK	GGEIEGTGSN	NGEELQMADD	TRLPMSRVVP	GKYHEGKGGEIEGTGSNGEELQMADDTRLPMSRVVPIPSSRLTPYRVVIIL	IIL
Ath-A	VIK	HEGGNNG	RGSNDDDDELI	DPDMPMMDE	GRQPLSRKLP	VIKHEGGNNGRGSNDDDELDDPDMPMMDEGRQPLSRKLPIRSSRINPYRML1LC	IIC
S0542	QVA	NKYPEAR	GGDMEGTGS	<b>IGEDIQMVDD</b>	ARLPLSRIVE	QVANKYPEARGGDMEGTGSNGEDIQMVDDARLPLSRIVPIPSNQLNLYRIVIIL	IIL
Ath-B	PV	STQAASERGG	VDIDASTDII	ADEALLNDE	ARQLLSRKVS	PVSTQAASERGGVDIDASTDILADEALLNDEARQLLSRKVSIPSSRINPYRMVIML	IMI
Cel-Al	PAT	TKV	EREAEIPPE(	QMEDKPAPD	ASOPLSTIIP	. TKVEREAEIPPEQQMEDKPAPDASQPLSTIIPIPKSRLAPYRTVIIM	IIM
Cel-A2							
D48636	PMTNGT	SIAPSEGRGV	GDIDASTDY	NMEDALLNDE	TROPLSRKVP	PMTNGTSIAPSEGRGVGDIDASTDYNMEDALLNDETRQPLSRKVPLPSSRINPYRMVIVL	IVL
					,		1
		310	320	330	340	350	360
RSW1	RLIILC	FFLQYRTTHP	VKNAYPLWL	<b>ISVICEIWFA</b>	FSWLLDQFPK	RLIILCFFLQYRTTHPVKNAYPLWLTSVICEIWFAFSWLLDQFPKWYPINRETYLDRLAI	LAI
Ath-A	RLAILG	LFFHYRILHP	VNDAYGLWL	<b>FSVICEIWFA</b>	VSWILDQFPK	RLAILGLFFHYRILHPVNDAYGLWLTSVICEIWFAVSWILDQFPKWYPIERETYLDRLSL	$\Gamma$ S $\Gamma$
S0542	RLIILM	FFFQYRVTHP	VRDAYGLWL	VSVICEIWLP	LSWLLDQFPK	RLIILMFFFQYRVTHPVRDAYGLWLVSVICEIWLPLSWLLDQFPKWYPINRETYLDRLAL	LAL
Ath-B	RLVILC	LFLHYRITNP	VPNAFALWL	VSVICEIWFA	LSWILDQFPK	RLVILCLFLHYRITNPVPNAFALWLVSVICEIWFALSWILDQFPKWFPVNRETYLDRLAL	LAL
Cel-A1	RLIILG	LFFHYRVTNP	VDSAFGLWL!	<b>ISVICEIWFA</b>	FSWVLDQFPK	RLIILGLFFHYRVTNPVDSAFGLWLTSVICEIWFAFSWVLDQFPKWYPVNRETYIDRLSA	LSA
Cel-A2							
<b>D48636</b>	RLVVLS	IFLHYRITNP	VRNAYPLWL	LSVICEIWFA	LSWILDQFPK	RLVVLSIFLHYRITNPVRNAYPLWLLSVICEIWFALSWILDQFPKWFPINRETYLDRLAL	LAL

FIGURE 9 (CONT III)

RSW1	370 380 390 400 410 420 RYDRDGEPSQLVPVDVFVSTVDPLKEPPLVTANTVLSILSVDYPVDKVACYVSDDGSAML
Ath-A S0542	RYEKEGKPSGLAPVDVFVSTVDPLKEPPLITANTVLSILAVDYPVDKVACYVSDDGAAML RYDREGEPSQLAPIDVFVSTVDPLKEPPLITANTVLSILAVDYPVDKVSCYVSDDGSAML
Ath-B	RLVILCLFLHYRITNPVPNAFALWLVSVICEIWFALSWILDQFPKWFPVNRETYLDRLAL
Cel-A1	RYEREGEPDELAAVDFFVSTVDPLKEPPLITANTVLS1LALDYPVDKVSCYISDDGAAML
D48636	RYDREGEPSQLAAVDIFVSTVDPMKEPPLVTANTVLSILAVDYPVDKVSCYVSDDGAAML
	430 440 450 460 470 480
RSW1	TFESLSETAEFAKKWVPFCKKFNIEPRAPEFYFAQKIDYLKDKIQPSFVKERRAMKREYE
Ath-A	<b>TFEALSDTAEFARKWVPFCKKFNIEPRAPEWYFSQKMDYLKNKVHPAFVRERRAMKRDYE</b>
S0542	TFEALSETAEFARKWVPFCKKHNIEPRAPEFYFAQKIDYLKDKIQPSFVKERRAMKREYE
Ath-B	SFESLAETSEFARKWVPFCKKYSIEPRAPEWYFAAKIDYLKDKVQTSFVKDRRAMKREYE
Cel-A1	TFESLVETADFARKWVPFCKKFSIEPRAPEFYFSQKIDYLKDKVQPSFVKERRAMKRDYE
Cel-A2	RRWVPFCKKHNVEPRAPEFYFNEKIDYLKDKVHPSFVKERRAMKREYE
D48636	TFDALAETSEFARKWVPFVKKYNIEPRAPEWYFSQKIDYLKDKVHPSFVKDRRAMKREYE

FIGURE 9 (CONT IV)

540 009 LVYVSREKRPGYQHHKKAGAENALVRVSAVLTNAPFILNLDCDHYINNSKAMREAMCFLM **EFKVRINALVAKAQKIPEEGWTMQDGTPWPGNNTRDHPGMIQVFLGHSGGLDTDGNELPR EFKVKINALVATAQKVPEEGWTMQDGTPWPGNNVRDHPGMIQVFLGHSGVRDTDGNELPR EFKVRINALVAKAQKVPEEGWTMADGTAWPGNNPRDHPGMIQVFLGHSGGLDTDGNELPR EFKIRINALVSKALKCPEEGWVMQDGTPWPGNNTGDHPGMIQVFLGQNGGLDAEGNELPR EYKIRINALVAKAQKTPDEGWTMQDGTSWPGNNPRDHPGMIQVFLGYSGARDIEGNELPR EFKVRINALVAKAQKKPEEGWVMQDGTPWPGNNTRDHPGMIQVYLGSAGALDVDGKELPR** EFKVRINGLVAKAQKVPEEGWIMQDGTPWPGNNTRDHPGMIQVFLGHSGGLDTEGNELPR LIYVSREKRPGFQHHKKAGAMNALIRVSAVLTNGAYLLNVDCDHYFNNSKAIKEAMCFMM LVYVSREKRPGFDHHKKAGAMNSLIRVSAVLSNAPYLLNVDCDHYINNSKAIRESMCFMM LVYVSREKRPGFQHHKKAGAMNALVRVSAVLTNGPFILNLDCDHYINNSKALREAMCFLM LVYVSREKRPGYQHHKKAGAENALVRVSAVLTNAPFILNLDCDHYVNNSKAVREAMCFLM 530 520 580 510 570 LVYVSREKRPGFQHHKKAGAMNALIRVSAV 500 560 490 550 Cel-A2 Cel-A1 Cel-A2 048636 Cel-A1 S0542 Ath-A Ath-B Ath-A Ath-B S0542 **RSW1** RSW1

FIGURE 9 (CONT V)

**D48636** 

LVYVSREKRPGFQHHKKAGAMNALVRVSAVLTNGQYMLNLDCDHYINNSKALREAMCFLM

	610	620	630	640	650	099
RSW1	DPAIGKKCCYVQFPQRFDGIDLHDRYANRNIVFFDINMKGLDGIQGPVYVGTGCCFNRQA	PORFDGIDLHDRY	(ANRNIVFFD)	NMKGLDGIQ	GPVYVGTGCCFN	RQA
Ath-A	DPQSGKKVCYVQFPQRFDGIDRHDRYSNRNVVFFDINMKGLDGIQGPIYVGTGCVFRKQA	?QRFDGIDRHDRY	SNRNVVFFDI	NMKGLDGIQ	GPIYVGTGCVFR	KQA
S0542						
Ath-B	DPNLGKQVCYVQFPQRFDGIDKNDRYANRNTVFFDINLRGLDGIQGPVYVGTGCVFNRTA	PORFDGIDKNDRY	(ANRNTVFFD)	NLRGLDGIQ	GPVYVGTGCVFN	RTA
Cel-A1	DPQVGRDVCYVQFPQRFDGIDRSDRYANRNTVFFDVNMKGLDGIQGPVYVGTGCVFNRQA	PORFDGIDRSDRY	<b>CANRNTVFFDV</b>	NWKGLDGIQ	GPVYVGTGCVFN	RQA
Ce1-A2	DPQFGKKLCYVQFPQRFDGIDRHDRYANRNVVFFDINMLGLDGLQGPVYVGTGCVFNRQA	PORFDGIDRHDRY	(ANRNVVFFD)	NMLGLDGLQ	GPVYVGTGCVFN	RQA
D48636	DPNLGRSVCYVQFPQRFDGIDRNDRYANRNTVFFDINLRGLDGIQGPVYVGTGCVFNRTA	PORFDGIDRNDRY	(ANRNTVFFD)	NLRGLDGIQ	GPVYVGTGCVFN	RTA
	670	089	069	700	710	72.0
1220		TO 500 0 7777 T T 140 C	WAAD DADAA	12.		4
KUMT	LIGIURVLIEEULEFNILVNOCCGORNAGNOONINIE.	FINITANDOCOST	Y Y Y Y Y Y Y Y Y Y Y Y Y Y Y Y Y Y Y			. NAR
Ath-A	LYGFDAPKKKKPPGKTCNCWPKWCCLCCGLRKKSKTKA	SKTCNCWPKWCCI	CCGLRKKSKT	ТКА	<b>М</b> К	KDKKT
S0542						
Ath-B	LYGYEPPIKVKHKK	KHKKPSLLSKLCGGSRKKNSKAKKESDK.	RKKNSKAKKES	3DK	KK	.KKSGR
Cel-A1	LYGYGPPSMPSFPK	SFPKSSSSSCSCCCPGKKEPKDPSELYRDA	CCPGKKEPKI	)PS	ELY	RDA
Cel-A2	LYGYDPPVSEKRPKMTCDCWPSWCCCCCGGSRKKSKKKGEKKGLLGGLLYGKKKKMMGKN	CMTCDCWPSWCCC	CCGGSRKKSK	CKKGEKKGLL	GGLLYGKKKKMM	GKN
D48636	LYGYEPPIKQKKKGSFLSSLCGGRKKASKSKKKSSDK	SSFLSSLCGGRKE	<b>CASKSKKKSSI</b>	)K	KKSNK	SNK

	730	740	750	160	770	780
RSW1	GINRSDSNAPLFNMEDIDEGFEGYDDERSILMSQRSVEKRFGQSPVFIAATFMEQGGIPP	<b>IEDIDEGFEGYD</b> I	ERSILMSQRS	VEKRFGQSPV	FIAATFMEQGC	IPP
Ath-A	NTKETSKQIHALENVDEGVIVPVSNVEKRSEATQLKLEKKFGQSPVFVASAVLQNGGVPR	IVDEGVIVPVSNV	TEKRSEATQLK	LEKKFGQSPV	FVASAVLQNG	VPR
S0542						
Ath-B	HTDS.TVPVFNLDDIEEGVEGAGFDDEKALLMSQMSLEKRFGQSAVFVASTLMENGGVPP	) I E E G V E G A G F D L	EKALLMSOMS	LEKRFGQSAVI	FVASTLMENGO	WPP.
Cel-Al	KREELDAAIFNLREIDNYDEYERSMLISQTSFEKTFGLSSVFIESTLMENGGVAE	IDN YDEY	ERSMLISQTS	FEKTFGLSSV	FIESTLMENG	VAE
Cel-A2	YVKKGSAPVFDLEEIEEGLEG. YEELEKSTLMSQKNFEKRFGQSPVFIASTLMENGGLPE	IEEGLEG. YEEI	EKSTLMSQKN	FEKRFGQSPV	FIASTLMENG	LPE
D48636	HVDS.AVPVFNLEDIEEGVEGAGFDDEKSLLMSQMSLEKRFGQSAAFVASTLMEYGGVPQ	) I E E G V E G A G F D L	EKSLLMSQMS	LEKRFGQSAA	FVASTLMEYGO	VPQ
	790	008	018	820	0.5	840
RSW1	TINPATLIKEAIHVISCGYEDKTEWGKEIGWIYGSVTEDILTGFKMHARGWISIYCNPPR	1SCGYEDKTEWG	KEIGWIYGSV	TEDILTGFKM	HARGWISIYC	PPR
Ath-A	NASPACLLREAIQVISCGYEDKTEWGKEIGWIYGSVTEDILTGFKMHCHGWRSVYCMPKR	ISCGYEDKTEWG	KEIGWIYGSV	TEDILTGFKM	HCHGWRSVYCN	IPKR
S0542						
Ath-B	SATPENFLKEAIHVISCGYEDKSDWGMEIGWIYGSVTEDILTGFKMHARGWRSIYCMPKL	ISCGYEDKSDWG	MEIGWIYGSV	TEDILTGFKM	HARGWRSIYC	IPKL
Cel-Al	SANPSTLIKEAIHVISCGYEEKTAWGKEIGWIYGSVTEDILTGFKMHCRGWRSIYCMPLR	ISCGYEEKTAWG	KEIGWIYGSV	TEDILTGFKM	HCRGWRSIYCN	IPLR
Cel-A2	GTNSTSLIKEAIHVISCGYEEKTEWGKEIGWIYGSVTEDILTGFKMHCRGWKSVYCVPKR	'ISCGYEEKTEWG	KEIGWIYGSV	TEDILIGFKM	HCRGWKSVYCV	'PKR
D48636	SATPESLLKEAIHVISCGYEDKTEWGTEIGWIYGSVTEDILTGFKMHARGWRSIYCMPKR	'ISCGYEDKTEWG	TEIGWIYGSV	TEDILTGFKM	HARGWRSIYCN	IPKR

FIGURE 9 (CONT VII)

	850	860	870	880	068	900
RSW1	PAFKGSAPINLSDRLNQVLRWALGSIEILLSRHCPIWYGYHG. RLRLLERIAYINTIVYP	NOVLRWALGSI	EILLSRHCPI	WYGYHG.	RLRLLERIAYINTIV	NP
Ath-A	AAFKGSAPINLSDRLHQVLRWALGSVEIFLSRHCPIWYGYGG.GLKWLERFSYINSVVYP	HQVLRWALGSV	EIFLSRHCPI	WYGYGG.	GLKWLERFSYINSVV	ΛYΡ
S0542						
Ath-B	PAFKGSAPINLSDRLNQVLRWALGSVEILFSRHCPIWYGYNG.RLKFLERFAYVNTTIYP	VOVLRWALGSV	EILFSRHCPI	WYGYNG	RLKFLERFAYVNTTI	IXP
Cel-A1	PAFKGSAPINLSDRL	HQVLRWALGSV	EIFLSRHCPL	WYGFGGG	NLSDRLHQVLRWALGSVEIFLSRHCPLWYGFGGGRLKWLQRLAYINTIVYP	VYP
Cel-A2	PAFKGSAPINLSDRLHQVLRWALGSVEIFLSRHCPLWYGYGG.KLKWLERLAYINTIVYP	<b>1QVLRWALGSV</b>	EIFLSRHCPL	WYGYGG.	KLKWLERLAYINTIV	NP
D48636	PAFKGSAPINLSDRL	<b>NOVLRWALGSV</b>	EILFSRHCPI	WYGYGG.	NLSDRLNQVLRWALGSVEILFSRHCPIWYGYGG.RLKFLERFAYINTTIYP	ΙΧЪ
	910	920	930	940	950	960
RSW1	ITSIPLIAYCILPAFCLITDRFIIPEISNYASIWFILLFISIAVTGILELRWSGVSIEDW	CLITDRFIIPE	ISNYASIWFI	LLFISIA	VTGILELRWSGVSIE	EDW
Ath-A	WTSLPLIVYCSLPAVCLLTGKFIVPEISNYAGILFMLMFISIAVTGILEMQWGGVGIDDW	CLLTGKFIVPE	ISNYAGILFM	LMFISIA	VTGILEMQWGGVGID	MOC
S0542						
Ath-B	ITSIPLLMYCTLLAVCLFTNQF1IPQISNIASIWFLSLFLSIFATGILEMRWSGVGIDEW	CLFTNQFIIPQ	ISNIASIWFL	SLFLSIF	ATGILEMRWSGVGIL	OEW
Cel-A1	FTSLPLIAYCSLPAICLLTGKF1IPTLSNLASVLFLGLFLSIIVTAVLELRWSGVSIEDL	CLLTGKFIIPT	LSNLASVLFL	GLFLSII	VTAVLELRWSGVSIE	EDL
Cel-A2	FTSIPLLAYCTIPAVCLLTGKF1IPTLSNLTSVWFLALFLSIIATGVLELRWSGVSIQDW	CLLTGKFIIPT	LSNLTSVWFL	ALFLSII	ATGVLELRWSGVSIQ	MOZ
D48636	LTSIPLLIYCVLPAI	CLLTGKFIIPE	ISNFASIWFI	SLFISIE	CVLPAICLLTGKF11PEISNFASIWF1SLF1S1FATG1LEMRWSGVG1DEW	OEW

18/21

1080 PPTTVLLVNLIGIVAGVSYAVNSGYQSWGPLFGKLFFALWVIAHLYPFLKGLLGRQNRTP PPTTLLIINIIGVIVGVSDAISNGYDSWGPLFGRLFFALWVIVHLYPFLKGMLGKQDKMP PPTTLLIVNLVGVVAGVSYAINSGYQSWGPLFGKLFFAFWVIVHLYPFLKGLMGRQNRTP PPTTLLIVNMVGVVAGFSDALNKGYEAWGPLFGKVFFSFWVILHLYPFLKGLMGRONRTP PPTTLIILNMVGVVAGVSDAINNGYGSWGPLFGKLFFAFWVILHLYPFLKGLMGRONRTP WRNEQFWVIGGASSHLFALFQGLLKVLAGVNTNFTVTSKAAD.DGAFSELYIFKWTTLLI WRNEQFWVIGGVSAHLFAVFQGLLKVLAGVDTNFTVTAKAAD.DTEFGELYLFKWTTLLI WRNEQFWVIGGISAHLFAVFQGLLKVLAGIDTNFTVTSKASDEDGDFAELYMFKWTTLLI WRNEQFWVIGGTSAHLFAVFQGLLKVLAGIDTNFTVTSKATDEDGDFAELYIFKWTALLI WRNEQFWVIGGVSAHLFAVFQGILKVLAGIDTNFTVTSKASDEDGDFAELYLFKWTTLLI WRNEOFWVIGGVSAHLFAVFQGFLKMLAGIDTNFTVTAKAAD.DADFGELYIVKWTTLLI 1070 1010 1000 1060 1050 990 980 1040 1030 970 Cel-A2 **D48636** Cel-A2 Cel-A1 Cel-A1 Ath-B Ath-B Ath-A Ath-A **S0542** S0542 RSW1 RSW1

FIGURE 9 (CONT IX)

**D48636** 

PPTTILIINLVGVVAGISYAINSGYQSWGPLFGKLFFAFWVIVHLYPFLKGLMGRQNRTP

TIVIVWSVLLASIFSLLWVRINPFVDANPNANNFNGKGGVF 1100 1090

TIIVVWSILLASILTLLWVRINPFVAK.GGPVLEICGLNCGN RSW1

Ath-A S0542

TIVVLWSVLLASVFSLVWVRINPFVSTADSTTVSQSCISIDC TIVVVWSVLLASIFSLLWVRIDPFTSRVTGPDILECGINC Cel-A1 Ath-B

TIVVLWSILLASIFSLVWVRIDPFLPKQTGPVLKQCGVEC TIVVVWAILLASIFSLLWVRIDPFTTRVTGPDTQTCGINC Cel-A2 D48636

FIGURE 9 (CONT X)

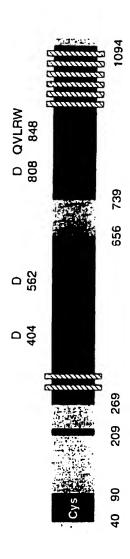
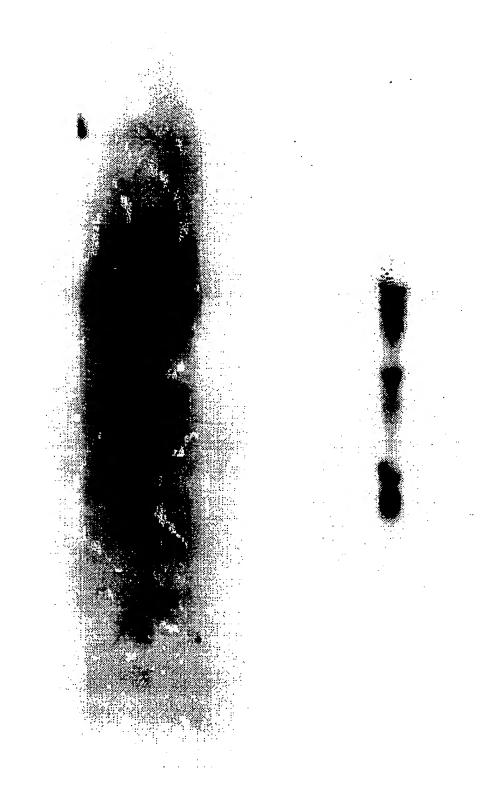


FIGURE 10



### INTERNATIONAL SEARCH REPORT

International Application No.

		PCT/AU 97/00402
A.	CLASSIFICATION OF SUBJECT MATTER	
Int Cl <sup>6</sup> :	C12N 15/54, 9/10	
According to	International Patent Classification (IPC) or to both national classification and	IPC
В.	FIELDS SEARCHED	
	umentation searched (classification system followed by classification symbols) nic Database Box below	
Documentation See Electron	n searched other than minimum documentation to the extent that such documents are inc nic Database Box below	cluded in the fields searched
WPAT. Med	n base consulted during the international search (name of data base and, where practical dline, ChemAbs, Genebank, Swiss Prot, EMBL ns: Cellulose Biosynthesis, Cellulose Synthase, Sequence ID# 2.	le, search terms used)
C.	DOCUMENTS CONSIDERED TO BE RELEVANT	
Category*	Citation of document, with indication, where appropriate, of the relevant pass	sages Relevant to claim No.
x x	WO 91/13988 (THE BOARD OF REGENTS, THE UNIVERSITY OF TEXAS SYSTEM), 19 September 1991 see whole document  WO 92/18631 (WEYERHAESUR COMPANY) 29 October 1992 see whole document	1-4, 14-16, 30-32 1-4, 14-16
x	WO 90/12098 (CETUS CORPORATION) 18 October 1990 see whole document	1-4, 14-16
	Further documents are listed in the continuation of Box C  X  See patent family	annex
* Special representation of the spec	After the international filing date or flict with the application but cited to theory underlying the invention cannot not be considered to involve an example the claimed invention cannot example; the claimed invention cannot inventive step when the document is other such documents, such to a person skilled in the art me patent family	
Date of the acti	Date of mailing of the international search  18 AUG	
	ing address of the ISA/AU INDUSTRIAL PROPERTY ORGANISATION  Authorized officer  Philippa Wyrdema	

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### INTERNATIONAL SEARCH REPORT

... ternational Application No.

PCT/AU 97/00402

Box 1	Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)
This Interna	ntional Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following
1.	Claims Nos.: because they relate to subject matter not required to be searched by this Authority, namely:
2. <u>X</u>	Claims Nos.:  because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:
3.	Claims Nos.:  because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a)
Box II	Observations where unity of invention is lacking (Continuation of item 2 of first sheet)
This Interna	ational Searching Authority found multiple inventions in this international application, as follows:
1.	As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims
2.	As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3.	As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:
4.	No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:
Remark on	Protest
	No protest accompanied the payment of additional search fees.

#### INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No. PCT/AU 97/00402

This Annex lists the known "A" publication level patent family members relating to the patent documents cited in the above-mentioned international search report. The Australian Patent Office is in no way liable for these particulars which are merely given for the purpose of information.

Patent Doo	cument Cited in Search Report			Patent	Family Member		
wo	9113988	AU	75569/91				
wo	9012098	AU	54373/90	CA	2014264	EP	471687
		IL	94053	NZ	233312	US	5268274
wo	9218631	US	5268274	NZ	233312	CA	2014264
		IL	94053	AU	54373/90	EP	471687

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